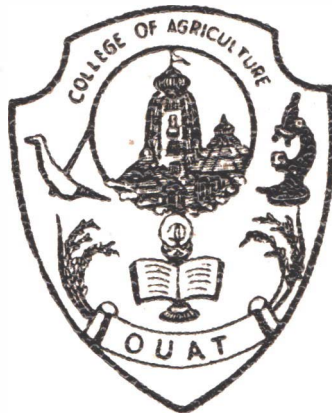


STUDIES ON INDEPENDENT AND COMBINED MUTAGENIC TREATMENT ON INDUCTION OF MUTATION IN GREENGRAM

A THESIS
SUBMITTED TO
THE ORISSA UNIVERSITY OF AGRICULTURE AND TECHNOLOGY
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FOR THE DEGREE OF

DOCTOR OF PHILOSOPHY
IN
PLANT BREEDING AND GENETICS

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CERTIFICATE - I

This is to certify that the thesis entitled "**Studies on independent and combined mutagenic treatment on induction of mutation in greengram Vigna radiata L. (Wilczek)**" submitted in partial fulfilment of the requirements for the award of the degree of **DOCTOR OF PHILOSOPHY** in **PLANT BREEDING AND GENETICS** of the Orissa University of Agriculture & Technology, Bhubaneswar, is a faithful record of bona fide research work carried out by **Mr. Laxmidhar Mishra**, under my guidance and supervision. No part of this thesis has been submitted for any other degree or diploma.

It is further certified that such help and sources of information as has been availed during the course of the investigation have been duly acknowledged.


(G.J.Patra)

CERTIFICATE - II

This is to certify that the thesis entitled "***Studies on independent and combined mutagenic treatment on induction of mutation in greengram, Vigna radiata L. (Wilczek)***" submitted by Mr. Laxmidhar Mishra to Orissa University of Agriculture & Technology, Bhubaneswar in partial fulfilment of the requirement for the degree of ***DOCTOR OF PHILOSOPHY in PLANT BREEDING AND GENETICS*** has been approved by the Advisory Committee and the External Examiner.

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Bhubaneswar
The 30th December, 1995.

L. D. Mishra
(L. D. MISHRA)

CONTENTS

Chapter	Title	Page
1.	INTRODUCTION	1
2.	REVIEW OF LITERATURE	6
2.1	Chemical mutagens and their mode of action	7
2.2	Methods of enhancing mutation frequency	11
2.3	Selection technique/procedures for quantitative characters in mutation breeding programme	17
2.4	Macro- and micromutations	22
2.5	Induce mutation in greengram	25
3.	MATERIAL AND METHODS	34
3.1	Materials used, variety and mutagen	34
3.2	Treatment methodology	35
3.3	M ₁ generation	38
3.4	M ₂ generation	40
3.5	M ₃ generation	42
3.6	M ₄ generation	42
3.7	M ₅ generation	43
3.8	Statistical methods	43
4.	EXPERIMENTAL RESULTS	55
4.1	M ₁ generation	55
4.2	M ₂ generation	66
4.2.1	Chlorophyll mutations in M ₂ generation	66
4.2.2	Morphological mutations in M ₂ generation	67
4.2.3	Total mutation frequencies in M ₂ generation	67

Chapter	Title	Page
4.3	Induce change in mean of the quantitative characters	71
4.4	Population variance	76
4.5	Correlation of M_1 parameters with macro mutation frequency in M_2 generation	80
4.6	Efficiency of single and combined mutagenic treatment	82
4.7	M_3 generation	85
4.8	M_4 generation	89
4.8.1	Mean of the quantitative characters	89
4.8.2	Variation, heritability and genetic advance	93
4.8.3	Interrelationship among characters	96
4.8.4	Path coefficient	100
4.8.5	Genetic divergence	103
4.8.6	Selection indices	115
4.9	M_5 generation	126
5.	DISCUSSION	128
-	Relationship between M_1 parameters with M_2 mutation frequency	131
-	Macro- and micromutation	132
-	Genetic variability, path, correlation	140
-	Genetic divergence	144
-	Grouping of mutant cultures	145
-	Selection indices	148
6.	SUMMARY AND CONCLUSION	151
	BIBLIOGRAPHY	159

LIST OF TABLES

Table	Title	Page
1.	Chemical mutagen used and their concentrations	35
2.	Details of mutagenic treatments used for the study	37
3.	ANOVA for Randomise block design	45
4.	ANOVA for compact family block design (Between treatment)	46
5.	ANOVA for compact family block design (Between progenies within treatment)	46
6.	ANOVA with expectations of mean sum of products under model	49
7.	Effect of single and combine treatments of mutagens on different M_1 treated populations Var. Dhauli	58
8.	Effect of single and combine treatments of mutagens on different M_1 treated populations Var. Nayagarh local	59
9.	Mean, Range of M_1 quantitative characters Var. Dhauli	64
10.	Mean, Range of M_1 quantitative characters Var. Nayagarh local	65
11.	Spectrum of different types of mutants Var. Dhauli	68
12.	Spectrum of different types of mutants Var. Nayagarh local	69
13.	Chlorophyll and morphological mutation frequencies	70
14.	Mean values of quantitative characters M_2 generation Var. Dhauli	74
15.	Mean values of quantitative characters M_2 generation Var. Nayagarh local	75
16.	Coefficient of variation of different characters, M_2 generation Var. Dhauli	78

Table	Title	Page
17.	Coefficient of variation of different characters, M ₂ generation Var. Nayagarh local	79
18.	Correlation between chlorophyll mutation with M ₂ parameters	81
19.	Correlation between M ₁ parameters with M ₂ population variance	81
20.	Relative efficiency of single and combine treatment of mutagens Var. Dhauri	83
21.	Relation efficiency of single and combine treatment of mutagens Var. Nayagarh local	84
22.	Mean single plant yield M ₃ generation Var. Dhauri	87
23.	Mean single plant yield M ₃ generation Var. Nayagarh local	88
24.	Mean values of quantitative characters M ₄ generation Var. Dhauri	91
25.	Mean values of quantitative characters M ₄ generation Var. Nayagarh local	92
26.	Genetic parameters of quantitative traits in mutant cultures Var. Dhauri	95
27.	Genetic parameters of quantitative traits in mutant cultures Var. Nayagarh local	95
28.	Phenotypic and Genotypic correlation among quantitative traits of mutant cultures Var. Dhauri	98
29.	Phenotypic and Genotypic correlation among quantitative traits of mutant cultures Var. Nayagarh local	99
30.	Direct and indirect effect of component traits on seed yield of mutant cultures Var. Dhauri	101
31.	Direct and indirect effect of component traits on seed yield of mutant cultures Var. Nayagarh local	102

Table	Title	Page
32.	Clustering pattern of mutant cultures of Dhauli on the basis of genotypic divergence	104
33.	Clustering pattern of mutant cultures of Nayagarh local on the basis of genotypic divergence	104
34.	Inter and intra cluster distance for Dhauli mutants	105
35.	Inter and intra cluster distance for Nayagarh local mutants	106
36.	Group means of 8 characters in different clusters (Dhauli)	110
37.	Group means of 8 characters in different clusters (Nayagarh local)	110
38.	Relative contribution of different characters to genetic diversity among mutant culture	112
39.	Selection index values of mutant cultures of Dhauli on the basis selection criteria	117
40.	Selection index values of mutant cultures of Nayagarh local on the basis selection criteria	118
41.	Mutant cultures selected on the basis of different selection criteria with ranking of index values for each selection criteria	119
42.	Expected genetic gain in yield and efficiency of selection on the basis of different selection criteria in M_4 generation	121
43.	Yield of mutant cultures of Dhauli selection on the basis of different selected criteria	122
44.	Yield of mutant cultures of Nayagarh local selection on the basis of different selected criteria	123
45.	Performances of selected mutant cultures in M_5 generation and it's correlation with respective index values Var. Dhauli	124

46.	Performances of selected mutant cultures in M_5 generation and it's correlation with respective index values Var. Nayagarh local	125
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LIST OF FIGURES

Figure	Title	Page
1	Statistical distance among 47 micromutants including control of Dhauli	113(a)
2.	Statistical distance among 47 micromutants including control of Nayagarh local	113(b)
3.	Scalter of points representing Dhauli in $Z_1 - Z_2$ graph with D^2 clustering super imposed	114(a)
4.	Scalter of points representing Nayagarh local in $Z_1 - Z_2$ graph with D^2 clustering super imposed	114(b)

CHAPTER - I
INTRODUCTION

INTRODUCTION

Pulses, being rich in protein (17-25 %) are important ingredient in the Indian diet and meet the needs of the large section of the people of the country. These are considered as poor man's meat because it implies a meat substitute. Recent indications of positive correlation of arterio - sclerosis with diet rich in saturated fatty acid on one hand and reported decrease in blood cholesterol level with inclusion of pulses on the other led the growing realization for substituting a part of animal protein by vegetable protein (Chandra and Ali, 1986). Besides, pulses are endowed with unique properties of maintaining and restoring soil fertility through biological nitrogen fixation.

India is major greengram growing country of the world, which shares 55 and 45 per cent of the total area and production of the world respectively. Despite the importance, progress in production and productivity remains far from satisfactory. The yield remained constant for last 3 decades and thus, per capita availability of pulses has progressively declined to 45 g/day as against 70 g/day during 1960-61. The efforts done in research and development of pulse crops have been rather marginal vis-a-vis the problems confronting crop improvement. There is a requirement of elimination of constraints coming in the ways of improving pulse production.

Due to continuous cropping of these crops on marginal and submarginal land, natural selection rather than human selection

played an important role for its existence (Bahl et al., 1979). This had favoured the crop to retain many semi wild characters like indeterminate growth habit, bushy and spreading plant type, late and asynchronous maturity and shattering of pods. These characters helped the crop to enhance its fitness of survival under relatively more unfavourable conditions (Jain, 1975). This is why, genes for favourable expression of yield components are likely to be eroded.

The available evidence shows that grain legumes have high genetic potential for yield (Jain, 1972, 1975; Swaminathan 1973). Therefore, through appropriate breeding methodology either conventional and/or mutation and efficient selection procedure, the morphological framework of the plant can be reconstructed in such a way to get determinant type with better yield, erect and compact habit, early and synchronous flowering and photo insensitive variety.

Induction of mutation has become an important tool for plant breeders with availability of mutagens and expansion of knowledge and technique for inducing mutation. Induction of mutations by radiation and their significance was demonstrated by Muller (1927) in Drosophilla and Stadler (1928) in maize. After discovery of chemical mutagen by Auerbach (1943) and Oehlkers (1943) it was understood that there are certain amount of specificity in biological damage as compared to physical mutagens (Konzak, 1965). Among the chemical mutagens, alkylating agents has a special position in inducing mutation. Ethyl methane sulphonate (EMS), Nitroso guanidine (NG) and Sodium azide (SA) are now a days extensively used as they

induce more of point mutation with negligible biological or chromosomal damage. The chemical mutagens, in addition to, are more powerful in terms of high mutation rate, than ionizing radiation. Expectations that chemical mutagen might give something better, more desirable for plant breeders than physical mutagens has led to the widespread use of chemical mutagens (Konzak et al., 1965 and Salnikova, 1991).

The specificity of mutagen for a particular type of genetic alteration, the conclusion seems to be that any specificity, if it exists, is of much little significance that it can not determine the choice of mutagen. In general, it would seem advisable to use several mutagens for a spectrum of mutations as wide as possible. (Nilan, 1972). Pulse crops differ in their sensitivity to mutagens and so do varieties within species. Different chemicals have different action on genetic materials, this prompted the mutational workers to go for combine application of mutagens either physical/chemical or chemical/chemicals to enhance the mutation frequency (Gupta, 1982; Grover and Virk, 1984; Rao et al., 1984). But then the precise control over mutation spectrum is yet to be achieved.

Induction of mutation can bring about changes both in qualitative and quantitative traits of different crop plants. This creates new variability and opens scope for selection (Gaul, 1961, 1964; Gustaffson, 1969). Mutations that alter the morphological characters of mungbean have been reported by Dahiya, 1973; Grover and Tejpaul, 1979; Grafia et al., 1983; Yadav and Singh, 1983; Grover and Virk, 1984; Pande and Raghubansi 1988; Khan 1989. These mutants alter the expression

of characters such as leaf type, leaf number, tallness, dwarfness, pollen sterility and so many characters.

Another group of mutations called micromutation cause minute variations in size and structure of quantitatively inherited characters have been identified in greengram. (Dahiya, 1973, 1978; Singh and Chaturvedi, 1981; Kundu and Singh 1982; Khan 1982, 1983, 1988, 1989 and so many). These micromutations are more useful to Plant Breeders than macromutation (Gregory, 1956). Induced micromutations are generally considered to be less associated with defective changes and hence, more readily exploited in breeding programme. These mutants are easily adopted to the ecological and environmental situation in which the parental genotypes are normally grown. The occurrence of micromutations have been demonstrated clearly to be of practical value in improving the yield potentials of established and adopted varieties (Khan, 1988; Sinha and Mohapatra, 1990). Therefore, mutational approach is increasingly directed towards the induction of micromutations.

The occurrence of mutation frequency is normally low. Therefore, probability of isolating mutants of economic interest depends on the ability of the breeder to select mutants from a large number of normal looking populations. Success of handling the materials depends upon the early elimination of non-mutant type which can be done by proper selection procedures. Further, the enhancement of mutation frequency, in turn, depends upon the availability of basic information on the mutagenic sensitivity of genotypes, use of

effectiveness and efficient mutagens and the vigour of diplontic and haplontic seives operating in the plant. The condition under which, a particular chemical mutagen induce different kinds of genetic changes should be determined, so that action of the mutagen can be regulated. Efficient and effective method of identification and selection of desirable micro-mutants need to be developed which otherwise will reduce the work load. It is reported by Tickoo and Jain, 1980; Khan, 1985; Khan and Hashim, 1979 that polygenic variation induced by mutagen could be fixed by selection in the subsequent generations and improvement of various desirable traits could be achieved.

Basing on the above considerations, the present investigation "Studies on independent and combined mutagenic treatment on induction of mutation in greengram *Vigna radiata* L. (Wilczek)" was undertaken with the objective to :

- (1) find the effective dose of mutagen and mutagenic combinations for inducing a wide spectrum and frequency of macro and micromutations,
- (2) find out the response of different mutagenic treatments in creating variability on different characters including yield in different varieties,
- (3) develop efficient selection methodology to select desirable micromutations, and
- (4) develop mutant lines with better yield performance *per se* and other desirable characters.

It is envisaged that the result of this investigation would be useful in isolating superior mutant cultures of greengram for future use.

CHAPTER - II

REVIEW OF LITERATURE

REVIEW OF LITERATURE

Induction of mutation by irradiation and their significance was demonstrated by Mullar (1927) in Drosophilla, Stadler (1928 a,b) in maize and barley and during 1929 by Goodspeed et al. in Nicotiana. This marked a new era both in fundamental and applied genetic research. The discovery of chemical mutagens during early forties, set another mile-stone in the history of mutational research. Auerbach (1943), Ochlkens (1943) and Rapoport (1946) had clearly demonstrated the powerful existence of chemical mutagens to induce variability.

The usefulness of any mutagen depends not only on its effectiveness but also on the efficiency of mutagen. Efficient mutagen may be defined as the production of desirable changes not associated with deleterious effects like gross chromosomal aberration, sterility, lethality, etc. (Konzak et al., 1965). Physical and chemical mutagens cause mutations in different ways. The use of physical mutagen in experimental biology posses certain limitations. Physical mutagen cause more of chromosomal aberrations and it's action is extremely random in DNA double helix, thus leading to high degree of meiotic and mitotic instability. The plant takes longer time to recover for which the initial growth, adoption and stability of the treated material is slow. Specifically this effect is more intense in

polygenically controlled characters. In addition to, easy availability is also another problem in the limited use of physical mutagens. After the discovery of chemical mutagens it has been found that chemical mutagens are more powerful in terms of high mutation rate than ionizing radiation. According to Gaul (1965) micromutations are more useful in plant breeding because : (1) they might occur much more frequently than macro mutants and (2) they might not so often result in reduced vitality as do most macromutations, because the change in physiological behaviour was supposed to be less drastic.

2.1 CHEMICAL MUTAGENS AND THEIR MODE OF ACTION

The different groups of chemicals identified were base analogues, alkylating agent, purine, hydroxylamine, nitrous acid, acridine, hydrazine, bisulphate, azide, antibiotic and radiosomic substances. These groups of mutagens included several chemicals and their derivatives. The mode of action of various types of chemicals is different. Alkylating agent is the outstanding and effective one in crop improvement programme. The various alkylating agents are sulphur mustard, nitrogen mustard, exoxides, ethylene imines alkyl alkane sulphonates, dialkyl sulphates, beta lactones, diazo compounds and nitroso compound.

The other type of mutagen used in crop improvement programme is azide group. Azide has the desired characteristics of producing primarily, if not solely, point mutations with no or little evidence of chromosomal aberrations as are commonly found with ionizing radiations. Although a point mutation, it is relatively safe, nonpersistent (Smith, 1956) and inexpensive

(Kleinhof et al. 1974; Sander et al. 1978 and Sato & Gaul, 1967). The lack of gross chromosomal changes could make azide more specific and more efficient use in plant improvement programme. Besides, sodium azide was proved to be a potent mutant in barley (Konzak et al. 1965; Nilan et al. 1973) and in rice (Hasegawa et al. 1980).

2.1.1 Ethyl methane sulphonate (EMS) $\text{CH}_3\cdot\text{SO}_2\cdot\text{O}\cdot\text{C}_2\text{H}_5$)

Bautz and Freese, (1960) and Freese, (1963) reported that this mutagen is well known for its high mutagenic efficiency due to alkylation. It induces more of gene mutations and less of chromosomal breaks. This react particularly to base guanine. EMS react with three of the four bases in the order of guanine, adenine, cytosine. This can react either with phosphate group or purine base especially with guanine of the DNA molecule (Krieg, 1963). The reaction of guanine base associated with ethylation of N-7 position is reported to be important pathway. Bautz and Freese (1960) reported that mutagenic action of EMS is mainly due to removal of guanine. The important element of EMS in the member of functional groups, i.e., mono, bi or poly functional. The bifunctional alkylating agent's toxicity is greater than the other two. EMS is very reactive agent and can react with water indicating the use of freshly prepared solution. The biological effects induced by EMS is usually measured in terms of percentage of lethality, seedling injury, pollen sterility in M_1 generation (Konzak et al. 1975 and Swaminathan et al. 1968). Due to loosening of bond between the base and sugar phosphatic chain, the "Apurinic gaps" is created, which if filled up, produces transition and

transversion mutants. If not filled up, it may result in frame-shift mutations (Lawley and Brookes, 1963).

Efficient use of EMS to induce higher frequencies of mutation over radiation has been observed by many mutation workers Swaminathan (1969) in wheat; Nerkar (1977) in lathyrus, and Lu et al. (1986) in rice. Acceleration of the frequency of mutation in mungbean has been accomplished by treating the seeds, with EMS (Santosh, (1969); Dahiya, (1973); Singh and Chaturvedi, (1980); Yadav and Singh, (1983); Grover and Virk (1984); Khan (1984); Sainy et al. (1989); Sahoo (1994) and many others.

2.1.2 Sodium azide (SA)

Sodium azide (NaN_3), though an invitro metabolite was found in the 1970s by Nilan et al. (1973) to be highly effective in producing mutations in barley, pea, several other plants and microbial species. This being an inhibitor of catalase and peroxide amylase was known to be effective under certain treatment conditions (Wyss et al. 1948). The potentially high mutagenecity of azide was discussed by Spance (1965) in barley. It produces high frequencies of gene mutations with negligible amount of chromosomal breaks (Konzak, et al. 1972; Nilan et al. 1973; Kleinhofs et al. 1974). Sodium azide is reported to be most efficient and effective in inducing Chlorophyll-deficient, morphological and bio-chemical mutants in barley (Konzak et al. (1965); Kleinhofs et al. (1974); Konzak et al. (1975) and Nilan et al. (1976). It is further reported by Nilan et al. (1976) that azide treatment resulted some delay in germination and low biological damage in M_1 generation. Kleinhofs, et al. (1974,

1978), reported that azide mutagenesis was due to inhibition of catalase, peroxide and cytochrome oxidase.

Azide is relatively safe and non-persistent and inexpensive (Smith 1969, 1966) and is very effective in acid medium while, less effective in alkali medium. Nilan, et al. 1973; Kleinhofs, et al. 1974 and Nilan, et al. 1973 suggested that by maintaining the pH of the azide solution at 3.0 and adjusting metabolic state of the seeds, mutations frequency induced by it would be equal to that of EMS treatment.

The exact mode of action as a mutagen is not known (Nilan et al., 1975), base substitution has been postulated to explain the mutagenic effect of sodium azide (Kleinhofs, et al. 1974). Hydrogen ions at pH 2 to 8 depurinates the DNA in-vitro (Sideris and Argyrakis, 1974) and a preferential liberation of adenine over guanine is observed.

Induction of micromutations by sodium azide has been reported by Shaikh et al. (1982); Iqbal and Khan (1989); Khan et al. (1989), Mahna et al. (1990) in different crops. Induction of micromutation in quantitative traits by SA has been reported by Krausse (1983); Mohapatra et al. (1983) Samal (1986); Iqbal and Khan (1989); Songwan and Singh (1990); Subudhi et al. (1991); Mishra (1992) and Sahoo (1994).

2.1.3 Nitroso guanidine (NG)

N-Methyl-N-nitro-Nitroso guanidine (NG), commonly called as Nitroso guanidine ($\text{CH}_3 \cdot \text{NHO} \cdot \text{CNH} \cdot \text{NH} \cdot \text{NO}_2$) is an alkylating agent containing methyl group. The mutagenic properties of NG was first reported by Rapoport (1946). During sixties, the mutagenic activity of NG was established in various test

materials (Eisenstark, et al. 1965; Gillham, 1965; Rapoport et al., 1966; Cerda - Olmedo and Hanawatt, 1967; Baker and Tessman, 1968; Browning, 1969).

Eisenstark, et al. (1965) and Baker and Tessman (1968) reported that NG produces equal proportion of GC to AT and AT to GC transition. The methyl group of NG react with Purine base and prodces more of methylated guanine than methylated adenine (Singer, et al., 1968). dong (1982) working in barley observed that NG induced structural, chromosomal and chromatic rearrangement and the effect increases with concentration. Floria, et al. (1982) reported that NG induced more chromosomal aberrations than EMS.

There are only few reports (Raveendran et al. 1982 and Samal 1986) on induction of micromutations in quantitative traits using NG treatment.

NG treatment was reported to have drastic effect on germination, survival, seedling growth parameters and pollen and seed fertility in M_1 generation. (Kempanna, et al., 1969; Sahoo, 1971; Levy and Anshri, 1973; Wibersbinski, 1984). Induction of micromutation by NG treatment in different crops have been reported by Sahoo and Samolo (1973); Swaminathan (1969); Kempanna et al. (1969); Prasad (1972); Uhlik (1973); Subramani et al. (1983); Vierling (1986); Mahasi and Sivasankar (1991); Mishra (1992).

2.2 METHODS OF ENHANCING MUTATION FREQUENCY

The mutation frequency is usually low which necessitate to select more efficient mutagens and methods involving suitable manipulation of extrinsic and intrinsic factors to

increase the frequency and spectrum of mutation. The techniques include - (1) manipulation of pre- and post-treatment conditions to lower lethality, thus enhancing sensitivity and mutation recovery, (2) evolving handling procedure to minimise haplontic or diplontic selections, (3) recurrent and sequential treatment, (4) combination treatment of mutagens and (5) different methods of treatment with radiation such as acute or chronic radiation.

Konzak et al. (1965) observed that washing of EMS treated seeds reduced the M_1 seedling injury and improves survival and fertility percentages. This was supported by Gaul et al. (1969) in barley.

Natarajan and Sivasankar (1965) found high mutation frequency by presoaking barley seeds for 16 hours before EMS treatment. Swaminathan (1969) stated that the enhancement in mutation frequency might be due to correspondence of treatment stage after 16 hours of presoaking with phase of DNA synthesis.

Joshi and Frey (1967) proposed recurrent treatment with different mutagens in successive generations as a possible method to increase the induced variability. they pointed out that alternate treatment of physical and chemical mutagens induce greater variability than recurrent treatment with the same mutagen in oat.

Dayal and Kaul (1969) observed that post treatment with GA reduced the damage of EMS treated and gamma irradiated barley seeds, in terms of germination, seedling injury and chromosomal abberation.

Sharma (1970) observed enlarged spectrum of viable mutations in gamma-rays and EMS treated wheat seeds, as compared to treatment of seeds with gamma-rays and EMS alone.

Sharma and Bansal (1970) reported high mutations frequency in barley when EMS treated seeds were dried back to 9.2 moisture content as compared to direct sowing after mutagen treatment.

Garina (1976) observed high chlorophyll mutation with increased period of presoaking followed by treatment. Treatment at G₁ phase increases the frequency of chromosomal aberration and decreased chlorophyll mutation as compared to treatment 'S' phase.

Babayan et al. (1983) presoaked the grains of wheat and barley at 60° to 80°C for 10 to 30 minutes before irradiation and select mutations in M₂ and M₃ generation. He also pointed out that heat treatment increased the frequency and range of mutation, in major quantitative traits governing yield.

2.2.1 Effect of combined treatments of mutagens

Combine treatments are reported to result the mutations more than the additive effect of single mutagen and such phenomenon are being describe as synergistic effect. A reason for the synergistic effect is that mutagen first applied may denature DNA and expose the protected metabolic site to the action of the second mutagen (Arnson et al. 1963). Alternatively the repair enzyme may render non-functional by the action of the second mutagen. which promotes the fixation of already included pro-mutational regimes (Sharma, 1970; Reddy et al. 1973). The synergism is also known due to

the effect of both enhanced inductions and more efficient fixation of primary mutational events. Rout (1979) treated greengram seeds with colchicine as pre-treatment before EMS and hydroxyl amine. This cause delay in flowering initiation by one week, enhanced pollen sterility and M_2 mutation frequency. Treatment with EMS increased the total frequency of mutation while the chlorophyll mutation frequency was increased in case of hydroxyl amine. Wide spectrum of agronomic mutation occurred in combination treatment.

Grover and Tejpaul (1979) observed that greengram seeds treated with maleic hydrazide and gamma-rays either single and in combination produced a number of micromutants.

Khan (1981) evaluated quantitative characters of greengram after treatment with gamma-rays and EMS either single and also in combination. The mean of the plant yield, number of pods per plant and 100-seed weight increased. Gamma-rays gave higher estimations of heritability and genetic advance for number of pods/plant and 100-seed weight. Similar results were observed for plant yield in case of combined treatment. The genetic variability induced could be exploited for selection of characters contributing to higher yield in later generations.

Singh et al. (1988) reported after treating greengram seeds with different concentrations of EMS, NMU and gamma-rays and their combination that all mutants were high yielders and high protein content.

Bahal and Gupta (1982) reported that combine treatment of gamma-rays and EMS produced higher mutation frequencies than individual treatments.

Bondopadhaya and Bose (1983) studied the cytological effects of greengram to single treatment with X-rays, EMS, HA and EI and pre and post irradiation treatments with these chemicals. It was reported that root tip showed chromosomal abnormalities like fragments, elumping of chromosome providing of metaphase early and unequal separation at anaphase originating from most treatments.

Krishnaswamy and Rathinam (1980) treated two cultivars of greengram with two levels of gamma-irradiation and EMS separately and jointly. It was observed that in M_2 generation, barring plant height, heritability in all other traits registered an increase under the mutagenic effect and enhancement of heritability, correlation between the characters underwent alterations under the mutagens.

Yadav (1987) treated greengram seeds to different doses of gamma-rays, EMS and their combinations. In M_3 most of the treatments resulted in more nodules in primary roots, than control. A high genetic coefficient of variation was observed for most of the characters studied except length of the primary roots which also showed lower heritability estimates. Nodules dry weight and nodules per plant were positively correlated with leaf area.

Rao et al. (1984) treating greengram seeds with EMS and gamma-rays found chlorophyll mutations in M_2 generation. It was observed that combine treatment of gamma-rays and EMS was superior to single mutagen treatment with respect to variation in quantitative characters.

Khalitkar and Bhargava (1986) observed different types of chlorophyll mutation frequencies when Hordeum seeds was treated with light and EMS in combination. It was indicated that different wave length of visible light also have modifying effect as mutation.

Das (1988) studied the genetic divergence among some induce mutants of greengram treating with EMS after Digitonin as pretreatment. Character association studies indicated that number of pods per plant, number of branches per plant and length of the pod were the three important contributing characters to genetic divergence.

Tripathy (1988) treated two varieties of greengram with different doses of gamma-rays EMS and their combinations. High doses of gamma-rays (30 kR) and combinations of 15 kR + 0.6 % EMS was found to be most suitable in inducing high frequencies of mutation in quantitative characters. Combine treatment of lower doses of EMS and gamma-rays was found suitable in inducing yield attributing characters for yield per se. Root weight, root length, hypocotyl length were found to be better parameters of M_1 in predicting M_2 frequencies.

Ghosh (1989) also continued the above study in M_4 and M_5 generations and concluded that combinations of lower doses of EMS and gamma-rays were better in inducing micro-mutational changes in greengram. There were indications for possibilities of existence of dose specificity for different quantitative characters.

Ignacimuthu et al. (1989) treated three species of Vigna with different doses of EMS, gamma-rays and their combinations.

It was revealed that the rate of induce variation in whole seed protein was narrow within the species, but within each mutant lines the range was broad. The whole seed protein showed genotype environment interaction both in control and mutant genotype.

Mahisi and Sivasnakar (1991) studied the combined effects of mutagens in cowpea. It was concluded that EMS gamma-rays produced polygenic mutation both in plants and hybrids with increased variance in polygenic traits. Combine treatment could produced additional variability for different quantitative characters.

Singh and Yadav (1991) treated different concentrations and combinations of gamma-rays and EMS in greengram and found different types of plant height, branch pattern, leaf morphology, pod characters, male sterility in M_2 and M_3 generation. The dwarf mutant VRDP-1 is possible a new source of dwarfism. Besides, five types of mutants obtained which alter different plant characters.

2.3 SELECTION TECHNIQUE/PROCEDURES FOR QUANTITATIVE CHARACTERS IN MUTATION BREEDING PROGRAMME

Gregory (1956) studied induction of mutation on groundnut and selected M_2 families on yield basis and advanced the selected single plants from this selection to M_3 generation. In the similar way, M_3 families were evaluated and advanced to M_4 generation for further evaluation and selection of high yielding lines.

Oka et al. (1958) followed the bulk population method of selection for heading date and plant height in rice. Normal

looking plants in M_2 were bulked and advanced to M_3 generation. Similar procedure were followed in M_3 and M_4 generation. Selection of single plants were advanced to M_5 generation.

Papa et al. (1961) applied first selection pressure in M_3 for yield, seed weight, maturity, oil content and protein content of soyabean and found differential response on yield to different varieties. However, selection was more effective for highly heritable traits like maturity, seed weight, oil content and protein content.

Mish and Yamaguchi (1965) practised selection on normal looking plants in M_2 generation and advanced them to M_3 generation. He further advanced five top yielding families from each treatment to M_4 for yield evaluation.

Bhatia and Van der Veen (1965) while working in Arabidopsis thaliana, raised M_2 generation as treatment bulk and applied selection pressure with 4.5 % selection intensity in M_2 for early and late flowering plants. In M_3 2% plants were selected from each M_3 families and same procedure of selection was followed up to M_6 generations.

Gaul et al. (1969) raised M_3 generation from selected normal looking M_2 plants. Selection in M_3 and M_4 generation were based on higher yield. They observed that selection initiated in M_3 was more efficient than that of M_4 .

Ojomo and Chheda (1972) applied selection pressure on yield in M_2 to raise M_3 generation. Twelve high yielding M_3 families from each treatments of cowpea were evaluated for yield in M_4 generation and then selected mutants for high yield.

Jana and Rây (1973) practised selection for yield and yield attributing characters in M_2 . Progenies significantly different from control were selected. M_3 generation was raised from selected bulk seeds of M_2 . Mutants with more number of grains/panicle and panicle length were realised following selection in M_3 generation.

Gill et al. (1974) working on barley, raised M_3 generation from the random selected plants of M_2 . Further 5 % selection intensity was applied on M_3 for 100-seed weight. In M_4 and M_5 generation also same selection procedures were followed and high yielding lines for 100-seed weight were obtained.

Sharma (1977) selected M_2 and M_3 generation of lentil. The selection response observed in M_2 and M_3 generation was variable from character to character studied.

Tickoo and Jain (1980) observed high variance in M_2 than M_3 of greengram indicating effective selection in M_2 . Selection was also applied in M_3 and M_4 and the selected lines were evaluated on M_5 generation and twenty five mutant lines with higher yield than parents were isolated.

Sharma (1982) treated the barley with physical, chemical mutagens and their combinations. He selected M_2 families with no shift in mean yield but high variance and raised M_3 generation. Superior families from M_3 were further tested in M_4 . A mutant culture, DL 253 with high yield than parents was developed.

Bahl and Gupta (1982) treated greengram seeds with gamma-rays and EMS. Selection was applied in M_2 and the same in

later generations they were evaluated in M_5 and M_6 generation. Mutants showing higher pod number, early, synchronous maturity were obtained.

Kharkwal (1983) suggested the selection techniques for micromutations in M_2 families, those have increase coefficient of variability and increase and/or no change in the mean values. The families showing greater superiority over control were advanced to subsequent generation for further selection.

Zakri et al. (1983) selected from the M_2 bulk of soyabean on the basis of yield, maturity and lodging resistance. Evaluation in latter generations showed a number of lines having earliness, lodging resistance and high yield.

Khan (1984) studied the variability in seeds/pod, 100-seed weight, and yield/plant in M_1 , M_2 and M_3 generations of greengram. It was observed that change of variability was dependent on character, treatment, generation and variety used. Genotypic coefficient of variation, heritability and genetic advance were higher in M_3 than M_2 indicating selection in M_3 generation for better response.

Sharma (1986) emphasised selection in early generation for micromutation in lentil. He indicated that selection for micromutations could be carried out in M_2 on the basis of mean and variance. Selection efficiency for quantitative characters could be increased with maximum mutagenic damage in the M_1 .

Devkota (1987) treated ragi seeds with EMS, NG for induction of micromutations. Normal looking high yielding M_2 plant progenies were selected at 33.3 % selection intensity. Similarly 25 % and 40 % selection intensity was applied in M_3

and M_4 generations respectively. On the basis of multiple criterion of selection 10 high yielding clusters were selected in M_5 . Finally two mutant lines with high yielding potential were identified in M_6 generation.

Sinha and Bharati (1990) selected promising plants in M_2 generation of irradiated blackgram and grown as family rows in M_3 generation. Further selection was applied in M_3 and advanced to M_4 . Several mutant lines showing more pods/plant than parent were isolated.

Kulkarni et al. (1991) tested seeds of blackgram varieties with gamma-rays and MS. Selection in both positive and negative directions was initiated in M_2 generation. The selected plants from the families were advanced to M_3 . From the M_3 family mean, they concluded that selection of high yielding M_2 plants in high yielding M_2 family was found to be affected by giving high yielding lines.

Sharma et al. (1991) treated greengram seeds with gamma-rays and raised M_2 from normal looking M_1 plants. Selection was given at inter and intra family levels for various characters on the basis of high mean and coefficient of variability. The selected lines were further advanced to subsequent generations and finally a bold podded mutant was isolated.

Nagabhusanam and Prasad (1992) treated seeds of groundnut with SA at a concentration of 3 μ m. He observed genotypic differences in response to mutagenic treatments in M_1 , M_2 and M_3 generations for various characters. Further, a strong correlation of canopy diameter at 60 days and with pod and

kernel yield was found indicating it's suitability as a selection criteria in breeding programme.

2.4 MACRO- AND MICROMUTATION

Induction of mutation can bring change in both qualitative and quantitative characters (Gregory 1956, 1961). Efficient mutagenesis involves production of desirable changes often free from other associated undesirable ones. Baur (1924) utilised the term "Grossmutationen" for large mutation and "Kleinmutationen" for small mutations. However, after the work of Gaul (1961, 1964). Swaminathan (1963, 1966) and Gustafsson (1969) the term "Macromutation" was used for large mutations affecting qualitative characters and "Micromutation" for small mutations affecting quantitative characters. Gaul (1964, 1965) was of the opinion that macromutations could be recognised with certainty in a single plant, while micromutations could be detected in a group of plants either by eye estimation or by measurement.

Knapp during early fifties presented a clear suggestion on the utility of micromutation in plant breeding. The usefulness of micromutation in plant improvement was suggested by Gregory (1956, 1961). However, after the work of Brock (1965 & 1967) and Gaul (1963, 1965) it became a common practice to advance the normal looking M_2 plants to M_3 and apply the first dose of selection not earlier than M_2 . Further, these worker were of the opinion that all sorts of quantitative characters are mutable. Though most of the reports suggest the induction of polygenic variability through mutation the opinions seem to differ regarding the direction of population mean. Therefore,

it is necessary to know, (a) the relative frequency of mutation with plus and minus effect, (b) factors affecting asymmetry of induced variability and (c) the magnitude of change and assume variability for response to selection.

The macromutations are associated with reduced fertility and variability due to pleiotropic effect of mutant genes or simultaneous mutations of closely linked genes in the background of genotypes. As reported by many workers, the use of micromutations in crop improvement brings change in plant ideotype, flowering time, leaf shape, leaf size, leaf number, seed size, protein content, nitrogen fixing ability, production of haploid, polyploid, male sterility, induction of resistance to diseases and pests, lodging, shattering, drought and other adverse conditions (Gaul, 1965, Oka et al. 1968, Scossiroli, 1965) Gregory (1955) and many other.

Change in quantitative traits are of great importance and have been utilised in cross breeding programme (Gaul et al. 1963), Scossiroli (1984, 1959). After the work of Gregory (1955) on the effect X-ray to induce genetic variability on yield in groundnut many workers like Brock and Latter (1961), Gaul et al. (1969), Bhatia and Swaminathan (1962), Swaminathan (1963), Gregory (1965), Brock (1965), Ehrenberg et al. (1965) and Scossiroli (1965), had published symmetric approach in the selection of micromutations. Many of them reported that induction of micromutation would enlarge the variability of the traits with or without affecting mean and therefore, it is required to assess the mean and variability of the treated population.

Bateman (1959), Brock (1965) observed that the incidence of polygenic mutations follow a trend opposite to that of previous selection of the parental genotype. On the contrary Oka, et al. (1958); Gaul (1965) and Reddy and Cherulu (1984) opined that the occurrence of micromutation was random in either direction. The induction of mutagen may bring symmetrical distribution, where plus and minus genes occurs in equal proportions. This finding was subsequently supported by Gregory (1956), Oka et al. (1958) in peanut; Gaul (1963) in barley; Yamaguchi (1964) in rice. Variation induced may be asymmetrical, where the plus and minus genes do not occur in equal frequency resulting a shift of mean in either direction. This hypothesis was supported by Brock and Latter (1961) in Subterrane, Ehrenberge et al. (1965) in rice and Scossiroli (1965) in wheat.

Since techniques and methods of estimating quantitative parameters have been refined, many workers have shown that mutation induced quantitative variations in various crop plants exhibit similar phenotypic expression to that released through hybridization. Oka, et al. (1958); Rawling et al. (1958); Gregory (1961) estimated that genetic variation induced through irradiation was approximately 50% of that of hybridization. Further, they proposed that induced mutation is as heritable and useful for either selection or crossing programme as that of hybridization.

During the last two decades several studies on induction of mutations on quantitative traits have been reported in pulse crops like, blackgram - Kundu 1980; Kundu & Singh 1982;

Kulshreetha and Singh 1983; Samal 1986; Iqubal and Khan 1989; Sinha and Bharati 1990; in greengram - Rajput (1974); Tickoo and Jain (1980); Bahl and Gupta (1982); Khan (1984, 1985); Songwan, (1988); Pea (Sharma, 1986), Lentil (Sharma, 1977, 1986; Ravi and Singh 1979); bengalgram (Ojomo and Chheda 1972, Kharkwal, 1981).

2.5 INDUCED MUTATIONS IN GREENGRAM

2.5.1 Germination

Sahoo and Samalo (1973) working on greengram applied MH and Colchicine as pre-treatment before EMS and found that there was reduction in germination and survival percentage. Colchicine as pre-treatment increased the pollen sterility in M_1 . With increase in pollen sterility, there was corresponding increase in mutation frequency in M_2 .

Singh and Chaturvedi (1980) treated greengram seeds with different concentrations of EMS, NMU, both single and in combination. The study revealed that combine treatments are most effective in inducing mutation than single application of mutagen. The seedling lethality was less in EMS treated M_1 populations.

Chandra and Tiwari (1979) and Subramani et al. (1983) studied the effects of gamma-irradiation on three species of Vigna in M_1 , M_2 and M_3 generations. The germination, survival and growth rate decreased with increase of dose level. In M_2 many chlorophyll mutants were noticed and viable morphological mutants were isolated for further study.

Mohapatra et al. (1987) reported that when two greengram varieties were subjected to different dosage of

gamma-irradiation, in both the varieties, M_1 survival percentage was significantly lower than the control. Further, there was a decrease in plant height. The length of the pod showed differential behaviour in both the varieties and the number of branches per plant was increased.

Ignacimuthu et al. (1989) treated the varieties of greengram seeds with different concentrations of gamma-rays, and EMS alone and in combinations. Mutagenesis lowered dehydrogenase in the M_1 and M_2 in all the species. Dose dependent decrease were noticed in seedling emergence, seedling height, survival and pollen sterility. The spectrum and frequency of chlorophyll mutations was increased with dosage.

2.5.2 Variation in quantitative characters

Dahiya (1973) treated two moong varieties with gamma-rays. The irradiation treatment was effective in inducing considerable variation for number of characters of economic value, i.e., yield/plant, grain size, number of pods per plant, days to flowering and protein content.

Mushataque and Rajput (1974) treated the seeds of greengram at dose level ranging from 10 to 40 kR and a study was made on the effects of gamma-rays on flowering time, plant height, pods per plant, pod length, grains per pod, 100-seed weight and seed yield.

In M_2 a shift in mean values of irradiated population for all polygenic traits occurred in a positive and negative direction except mean of the pod length which remained unchanged. The coefficient of variability was increased for all the traits studied. The additional variability was reported to

be genetic, which would provide opportunity in the improvement of greengram by selection.

Prasad (1976) isolated four mutants following EMS treatments in greengram which under high rainfall conditions gave convincingly better yield over the parents. It was further suggested that number of pods and growth pattern in greengram can be achieved by induction of mutation.

Khan and Hashim (1979) studied the mutagenic efficiency and effectiveness of gamma-rays, EMS and hydrazine hydrate in greengram and observed that mutagenic efficiency and effectiveness were higher at lower doses of mutagen.

Samolo, et al. (1979) found that stability in the yield of mutants were associated with stability of yield contributing characters. The mutant population remains heterozygous for macromutation and provides population buffereing and flexibility.

Krishnaswamy and Rathnam (1980) studied the frequency and spectrum of chlorophyll and viable mutations in relation to type and dose of mutagen. they reported that chlorophyll mutants did not vary with the mutagen dose, while viable mutants exhibited a direct relationship.

Mishra, et al. (1980) treated greengram seeds with EMS and found that EMS induced agronomic mutants. Maturity in greengram seems to be controlled by two groups of genes; one group tending towards earliness, while the other towards lateness. Further, this study revealed that selfed seed have higher mutation frequency than the open pollinated seeds; indicating certain amount of out crossing in M_1 due to

induction of pollen sterility. Even in a self pollinated crop like greengram upto 2.63% of mutants could not be expressed due to induction of pollen sterility and outcrossing.

Singh and Chaturvedi (1981) treated the greengram seeds with aqueous solution and observed that mutants showed a combinations of higher yield potential, earliness and tolerance to drought. The mutants derived also showed better stability in yield performance.

Khan (1981) irradiated the greengram seeds to different dosages of gamma-rays and found that mutation rate increased slightly at lower dose of radiation. There was also shift in the mean value of polygenic traits both in positive and negative direction. The genetic variation and estimation of heritability did not appear to dose dependent and varied with trait to trait. Variability increased considerably in all the treatments was due to mutation of polygens governing quantitative characters.

Mishra et al. (1981) treated three varieties of greengram with EMS. The regression analysis of M_2 mutation frequency in M_1 parameters showed that pollen size can be safely taken as criterion for predicting M_2 mutation frequency.

Mondihagaladza et al. (1981) treated two varieties of greengram with EMS, EI, NMU and found that most of the mutants were diploid except one treated with 0.005% NMU, which was syndoploid ($2n = 24$).

Mishra and Samolo (1982) reported that the frequency of mutation in greengram in M_3 was less than M_2 generation suggesting selection in M_2 .

Khan (1982) reported that radiation to greengram seeds shifted the mean values in all the characters mostly in a positive direction in M_2 and M_3 generations. The range of variability increased considerably. The GCV, heritability and genetic advance were increased. The genetic variability was not linear with dose. Different doses responded differently to mutagenic treatment.

Khan (1983) studied the induced variability of greengram in M_1 , M_2 and M_3 generations after treatment with gamma-rays, EMS and HZ. The variability increased in almost all characters in M_1 generation. There was an increase in mean values after gamma-rays and EMS treatment, but HZ failed to show same response. Estimation of heritability, genetic advance was higher for 100-seed weight, followed by plant yield and seeds/pod in M_2 generation. Genotypic coefficient of variation, heritability and genetic advance increased more in M_3 as compared to M_2 indicating that significant gain could be achieved by selection in M_3 generation.

Krishnaswamy (1983) following treatment of greengram varieties with EMS and gamma-rays observed a differential response of varieties to mutagens in terms of frequency of chlorophyll and viable mutants.

Mohapatra et al. (1983) treated two varieties of greengram with gamma-rays, EMS, NMU and SA. It was noticed that chemical mutagen has less biological damage than physical mutagen.

Mohapatra et al. (1983) compared the spectrum of induction of genetic variability on greengram after treatment

with gamma-rays, EMS, NMU and SA. Study of interaction coefficient showed synergistic effects in M_1 parameters and additive effect in M_2 . Treatment with sodium azide was found to be more effective. The genetic advance and heritability showed some positive changes in yield attributing characters.

Rao et al. (1983) irradiated greengram seeds with gamma-rays and found that plant height, number of clusters/plant was reduced in M_1 generation. There was an apparent increase of those characters in M_2 . Number of pods per cluster increased significantly in both the generations. Seed per pod was reduced with increase in dose of irradiation. The range for mean of the various characters was shifted in a positive direction and variance was increased significantly.

Khan (1984) studied the induced variability in quantitative characters of greengram after treatment with gamma-rays, EMS and HZ in M_1 , M_2 and M_3 generations. The mean number of seeds per pod and 100-seed weight was decreased whereas, the plant yield did not show any particular trend. There was an increase in the mean values after gamma-irradiation and EMS treatments in the M_1 generation. However, HZ failed to show same response. The mean value was increased in M_2 generation. Estimation of heritability was higher for 100-seed weight followed by plant yield and seeds/pod in M_2 generation. In general, the genotypic coefficient of variation, heritability and genetic advance increased more in M_3 as compared to M_2 indicating that significant gain could possibly be achieved through selection in M_3 generation.

Kadapa (1985) irradiated var. China Mung with different doses of X-ray and obtained plants with non-shattering pods in M_2 . All the plants which reached maturity from the 92 M_2 seeds sown were erect, taller with non-shattering pods. These mutants had longer pods and more seeds/pod and gave higher yield. The mutant showed greater variability than the parents.

Mohapatra et al. (1987) studied the genetic divergence among 36 mutant lines of 6 standard varieties of greengram and grouped them in 7 clusters. The seeds per pod and 100-seed weight contributed maximum towards genetic divergence. The clustering pattern indicated that mutant lines derived from a single mutagenic treatment could be quite diversified and the lines for different mutagenic treatments could be quite close to each other genetically.

Rosaiah et al. (1987) treated two varieties of greengram with gamma-irradiation and obtained 15 micro-mutants. It was observed that wide variability was induced in N-fixation traits through irradiation. The different yield attributing traits showed high heritability and were positively correlated with seed yield.

Yadav (1987) treated seeds of var. PS-16 with different dosages of EMS, diethyl sulphate and gamma-rays single and in combination. In general, the mitotic index and seedling vigour was decreased with increasing mutagenic dose. The effects of mitotic arrest were evident in roots sooner than on the shoots. Mutagenic treatment increased the frequency and spectrum of chlorophyll mutations.

Yadav and Singh (1987) in greengram variety T-44 detected male sterility in a mutant in the R_2 induced by 40 kR gamma-rays. These mutants had narrow leaves and compact habit and were having C-13 racemes/plant (v.s. C-10 in the parent) was on the basis of segregation pattern, controlled by single recessive gene which showed possible pleiotropic effect or belonged to a group of closely linked genes.

Chabra and Singh (1988) treated greengram seeds with EMS and found pentafoliate leaf type plants and crossed them to normal trifoliate types. In F_1 all plants were trifoliate. In F_2 , there was a segregate of trifoliate to pentafoliate in ratio of 3:1.

Khan (1978) evaluated three quantitative characters in Mungbean after irradiation with gamma-rays and EMS, using .pl67 either single or in combination. The mean of number of pods, 100-seed weight and plant yield increased in all treated plants as compared to control. The total phenotypic variability increased, so also the values of phenotypic and genotypic coefficient of variation.

Mohapatra et al. (1988) compared the mutagenic effects of SA and NG to two varieties of greengram. Induction of sodium adize had a wider effect on large number of characters, while it had a limited effect to few loci in case of NG. Induction of desirable mutants like early maturity, compact inflorescence, large bold pod size could be achieved through SA and NG treatment. The relative growth rate and pollen sterility in M_1 could be reliable parameter for predicting mutation frequency.

Songwan, (1988) observed that mutagenic effectiveness of gamma-rays, EMS and SA in different cultivars of mungbean and found that effectiveness and efficiency of mutagen was not dependent on dose, species, cultivars and varies with character choosen.

Yadav and Singh (1988) obtained a mutant which exhibited synchrony in ripening, earlier maturity and 39 % increase in yield in M_2 generation after irradiating greengram by gamma-rays.

Pulivarathi and Mary (1989) irradiated two varieties of greengram and observed that variability both in M_2 and M_3 generations in many of the yield components. To increase yield, it was reported that selection should be made for pods/plant and 100-seed weight on variety Pusa-105 and pods/plant and pods/cluster in variety Neelalu.

Seth and Choudhury (1989) observed increase in mean values for branches/plant in all the genotypes in M_2 and M_3 after irradiation with gamma-rays. The dose 20 kR was found to be most effective.

Sahoo (1994), observed that EMS with thyroxine pretreatment was more effective in inducing macromutations than other treatments and there was no definite trend of action to this effect due to pre-soaking periods. He further observed that different variety responded differently to mutagens.

CHAPTER - III

MATERIALS AND METHODS

MATERIALS AND METHODS

The investigation, "Studies on independent and combined mutagenic treatments on induction of mutation in greengram Vigna radiata L. (Wilczek)" was undertaken in EB-II Section, Department of Plant Breeding and Genetics, College of Agriculture from 1990 summer to 1992 kharif. The materials used details of the methods followed and the observations, recorded are outlined in the following sections.

3.1 MATERIALS

This experimental materials included two varieties of greengram namely Dhauli, a variety developed through mutation, and Nayagarh local a local cultivar. These two varieties are widely grown in Orissa. The salient morphological characters of these varieties are as follows.

3.1.1 Dhauli

Dhaulti is a promising variety evolved through mutation breeding in the Department of Plant Breeding and Genetics. It grows up to 30-40 cm height in Rabi and 50-55 cm in kharif. It matures in 60-65 days and has shinning green seeds with an yield variation of 5-15 quintals per hectare. This variety possess good vegetative vigour, high protein content (18 %) and is tolerant to powdery mildew.

3.1.2 Nayagarh local

This is a local variety grown in Orissa throughout the year. It is a well adopted variety and can be grown well under moisture stress condition. It grows up to 60 cm height and matures in 70 days. It has yield variation of 5.0 to 10.0 quintals per hectare. It has good vegetative vigour. The seed size is comparatively smaller than Dhauli. The yield is moderate.

3.1.3 Mutagens used

The chemical mutagens used in this investigation for induction of mutation were Ethyl methane sulphonate (EMS), Sodium azide (SA), and Nitroso guanidine (NG). Both the single and combinations of these mutagens were taken in this study. The details of the chemicals and their concentrations are presented in Table 1.

Table 1. Chemical mutagen used and their concentrations.

Sl. No.	Chemical Name	Formula	Concentrations used
1.	Ethyl methane sulphonate (EMS)	$\text{CH}_3\text{.SO}_2\text{.O.C}_2\text{H}_5$	0.2 % and 0.4 %
2.	Sodium azide (SA)	Na_3N_3	100 ppm and 200 ppm
3.	N-Methyl-N-Nitroso guanidine(NG)	$\text{CH}_3\text{-NO} \begin{array}{c} \text{NH} \\ \text{NH} \end{array} \text{-NH.NO}_2$	100 ppm and 200 ppm

3.2 TREATMENT MECHODOLOGY

Genetically pure, fully matured, uniform size and dry seeds (moisture content \approx 10.54%) of two varieties of greengram were selected for chemical treatment. In each treatment, 300 healthy seeds were soaked in distilled water for 12 hours.

The seeds after pre-soaking were blotted gently under the folds of absorbant paper to remove excess moisture. Then the chemical treatments were administered in closed petridishes. In case of single chemical treatment, pre-soaked seeds were put in mutagen solution for 6 hours. In case of combined treatments, the first 3 hours treatment was given with the first chemical and then seeds were washed thoroughly for 2 hours and blotted. Then the same seeds were treated with second chemical for another 3 hours. Twelve different treatment combinations were taken for the study in addition to 6 single treatments and one control. The control seeds were also pre-soaked in distilled water for 12 hours before use. The treatment was carried out with room temperature ($21 \pm 1^{\circ}\text{C}$). The details of the treatments and period of treatment is presented in Table 2. Thus, 19 different treatments including one control were administered to each of the two varieties.

3.2.1 Preparation of mutagenic solution

The action of a chemical mutagen depends upon the conditions like atmospheric temperature, pH of the water used for dilution etc. In this study, EMS was prepared by mixing the chemical with appropriate amount of distilled water. The Nitrosoguanidine was first dissolved in a little quantity of ethyl alcohol and then diluted to the required percentage by adding appropriate amount of distilled water. The sodium azide solution was prepared by dissolving appropriate quantity of sodium azide with phosphate buffer.

The seeds of both the varieties were treated with freshly prepared mutagenic solution. In order to ensure sufficient

Table 2. Details of mutagenic treatments used for the study.

Sl. No.	Treatment Code	Chemicals and concentrations	Period of treatment (hours)
1.	E ₁	EMS (0.4 %)	6
2.	E ₂	EMS (0.2 %)	6
3.	S ₁	SA 200 ppm	6
4.	S ₂	SA 100 ppm	6
5.	N ₁	NG 200 ppm	6
6.	N ₂	NG 100 ppm	6
7.	E ₁ S ₁	EMS (0.4 %) + SA 200 ppm	First mutagen for 3 hrs. and second one for 3 hrs.
8.	E ₁ N ₁	EMS (0.4 %) + NG 200 ppm	-do-
9.	S ₁ N ₁	SA 200 ppm + NG 200 ppm	-do-
10.	E ₂ S ₂	EMS (0.2 %) + SA 100 ppm	-do-
11.	E ₂ N ₂	EMS (0.2 %) + NG 100 ppm	-do-
12.	S ₂ N ₂	SA 100 ppm + NG 100 ppm	-do-
13.	E ₁ S ₂	EMS (0.4 %) + SA 100 ppm	-do-
14.	E ₁ N ₂	EMS (0.4 %) + NG 100 ppm	-do-
15.	S ₁ N ₂	SA 200 ppm + NG 100 ppm	-do-
16.	E ₂ S ₁	EMS (0.2 %) + SA 200 ppm	-do-
17.	E ₂ N ₁	EMS (0.2 %) + NG 200 ppm	-do-
18.	S ₂ N ₁	SA 100 ppm + NG 200 ppm	-do-
19.	Control	Distilled water	6

exposure of mutagen for seeds, the quantum of mutagenic solution used was approximately in the ratio of 3 parts of solution by volume to one part of seed volume. The contents of the beaker were stirred intermittently (in every 10-15 minutes) during the treatment period to maintain uniform concentrations of the solution through out the beaker and thereby render uniform penetrations of the mutagens into the seeds. After completion of the required period of chemical treatment, the treated seeds were washed in running water for 1-2 hours in order to leach out the residual cellular mutagen present in seeds which would otherwise may cause damage to the germinated seeds.

Fifty seeds from each treatments were sown in earthen pot (25 seeds in one pot) for laboratory study of seedling behaviour, i.e., root length, shoot length, seedling height, etc. Two hundred seeds were sown in the field to raise M_1 generation. Fifty seeds were kept to study mitotic abnormalities.

3.3 M_1 GENERATION

3.3.1 Materials and experimental design

Immediately after treatment, the seeds were sown in the field on 2nd March 1990 to raise M_1 generation. Two separate trials were conducted in RBD for two varieties. All the seeds of 19 treatments (including control) were sown in lines with two replications. A basal fertilizer dose of N_{10} and P_{40} kg/ha was applied at the time of sowing. Rest N_{10} kg/ha was topdressed after 21 days of sowing. A spacing of 30 cm between rows and 10 cm within row was followed.

3.3.2 Observation recorded

Germination: Observation on germination was recorded both in field and laboratory after 8 days of sowing and was expressed as percentage of seeds sown.

Survival : Number of plants that survived up to maturity was counted and expressed as percentage of seeds sown.

Seedling height : Average height of 20 randomly chosen seedlings was observed at 15 days after sowing.

Plant height : It was observed at maturity. The plant height (cm) was measured from ground level to tip of the plant.

Pods/plant : Number of pods/plant were recorded from 20 randomly selected plants at physiological maturity and was averaged to get pods/plant.

Seeds/pod : Five fully developed and fertile pods from each of randomly selected 20 plants were counted and number of seeds/pod was calculated.

100-seed weight : Seeds of all 20 randomly selected plants were bulked and 100 seeds were randomly chosen and weighed to arrive at 100-seed weight expressed in gram.

Pollen sterility : One mature floral bud from each of randomly selected plants were taken and fixed in aceto alcohol (1:3) for 24 hours. Then it was preserved in 70 % alcohol. The anthers from each of the floral bud were squashed using aceto Carmine stain and then observed under microscope to study the fertile and sterile pollen grains. The deeply stained pollen grains of standard size were considered as fertile while faintly or non-stained undersized pollen grains were recorded as sterile.

$$\text{Pollen sterility (\%)} = \frac{\text{Number of sterile pollens}}{\text{Number of total pollens}} \times 100$$

Seed sterility : Five randomly chosen pods from each of 20 random selected plants were taken. The under developed or undeveloped seeds were considered as sterile while fully developed seeds were taken as fertile.

$$\text{Seed sterility (\%)} = \frac{\text{Number of sterile seeds}}{\text{Total number of seeds}} \times 100$$

3.3.3 Mutagenic effectiveness

The interaction effect of combination treatments on mutagenic frequency was calculated by the formula :

$$K = \frac{a + b}{(a) + (b)}$$

where (a) and (b) are the mutation frequencies induced by the two mutagens when applied singly, whereas (a + b) is the mutation frequency produced by the two mutagens in combination treatment and 'K' is the hypothetical interaction coefficient (Sharma, 1970). If the value of 'K' is one, interaction is additive. Synergistic interaction may alter the value in positive or negative direction.

Recovery index (RI) of single and combination treatment in M_1 generation was calculated by using the formula $\bar{X} \times S$, where \bar{X} was the mean seedling height and 'S' is the corresponding percentage of survival in the seed treatment.

3.4 M_2 GENERATION

3.4.1 Materials and experimental design

The seeds from two fruiting branches from all M_1 plants

were bulked treatment wise. The bulked seeds were used to raise M_2 generation. It was grown in randomised block design (RBD) with three replications. Two separate experiments for the two varieties were conducted. An inter-row spacing of 30 cm and intra-row spacing of 10 cm was adopted. The crop was grown on 10th August 1990. Recommended cultural practices were followed during the growing season.

3.4.2 Observations

With the objective of studying the direction and spectrum of micromutations induced by different mutagens and their combination treatments. The variability induced in the treatments were compared with control. For this, twenty numbers of normal looking plants (excluding the macro-mutants) were randomly chosen. Observations on plant height, pods/plant, seeds/pod, 100-seed weight and yield/plant were recorded as described under M_1 generation.

Yield/plant : The weight (g) of the seeds obtained from a single plant was recorded and yield/plant was computed.

From the observed value of these twenty randomly selected plants, the mean and coefficient of variation on quantitative traits were calculated replication wise. This was used for further analysis.

3.4.3 Macro-mutations

The different types of chlorophyll mutants were recorded daily from 5th to 15th day after sowing. They were grouped into following categories. Observation on different foliar mutants were identified at 5 to 45 days after sowing. The other morphological mutants were recorded during vegetative and .pl67

reproductive phase of the crop. The major agronomic mutants observed were tall, dwarf, early, late, long and short poded, bushy, more poded, profused branched mutants, etc.

3.5 M₃ GENERATION

3.5.1 Materials and experimental design

Twelve number of plants per treatment (20% of 60 M₂ plants per treatment) were selected on the basis of single plant yield for growing M₃ generation. Progenies from these selected plants of both the varieties were grown in compact family block design with 3 replications. The treatments were taken in main plot and progenies on the sub-plot. Two separate trials were conducted for both the varieties. The trials were sown on 12.2.91 with the normal package of practice as described earlier in this chapter.

3.5.2 Observations recorded

The plant yield for each of the progenies was considered by taking mean of the 10 randomly selected normal looking plants. The mean of the 10 plants were taken as progeny mean for further analysis in compact family block design.

The progenies giving more than 50 % higher yield than the superior progeny of the control were selected. In this way, 46 progenies were selected each for Dhauli and Nayagarh local. These selected mutant progenies were grown along with the control to raise M₄ generation.

3.6 M₄ GENERATION

3.6.1 Experimental design

Forty six numbers of selected progenies and one control were grown to raise M₄ generation of Dhauli and Nayagarh local.

The seeds of the selected progenies were grown in RBD with 3 replications. The crop was grown on 5th August 1991 with recommended package of practices as described earlier in this chapter.

3.6.2 Observation recorded

Observations were recorded on 50 % flowering, maturity (days), plant height (cm), pods/plant, seeds/pod, 100-seed weight (g) and plant yield (g) by taking 10 normal looking plants from each of the mutant cultures, per replication as per the procedure outlined in M_1 generation and the mean data were computed. Observation on maturity and 50 % flowering was considered on visual estimation.

3.7. M_5 GENERATION

The selected mutants of M_4 were grown in M_5 generation in a RBD design with three replications. Two separate trials were conducted for two varieties. It was grown on February 1992 with normal package of practices.

Observations on days to maturity were taken on eye estimation. From ten randomly chosen plants from each treatment from each replication observations were recorded for plant height, pods/plant, seeds/pod, 100-seed weight and single plant yield. The observations were taken as per procedure followed earlier.

3.8 STATISTICAL METHODS

3.8.1 M_1 Generation

The different statistical analysis were carried out for all the five generations. The data generated and the procedures followed are furnished in this chapter.

The relative efficiency of mutagenic treatments in inducing variation for different quantitative traits were estimated through analysis of mean, range and coefficient of variability (CV).

Variance : The estimate of variance provides a measure of variation with respect to a particular character available in the population treatments.

It is expressed as follows :

$$\delta^2 = \frac{\Sigma x^2 - \{(\Sigma x)^2/N\}}{N - 1}$$

where, δ^2 = Variance

X = Individual observation

N = Number of observations

Co-efficient of variation (CV) : Using the variance component the coefficient of variation was estimated among all the treated population in M_1 as follows :

$$CV = \frac{SD}{Mean} \times 100$$

where SD is the standard deviation.

3.8.2 Relationship between M_1 parameters and M_2 chlorophyll mutation frequencies.

Relationship of M_1 parameters with chlorophyll mutations, frequencies and variability in quantitative traits of M_2 generation were calculated by means of correlation coefficient between these two parameters.

Correlation coefficient (r)

$$r = \frac{\delta_{(xy)}}{(\delta_x^2 \cdot \delta_y^2)^{\frac{1}{2}}}$$

where, x and y are the M_1 parameters and M_2 macro-mutations respectively,

$$\delta^2_x = \text{Variance of } x$$

$$\delta^2_y = \text{Variance of } y$$

$$\delta_{xy} = \text{Co-variance of } x \text{ and } y.$$

3.8.3 M_2 Generation

Different macromutation (chlorophyll and morphological frequency in M_2 generation were calculated as follows :

$$\text{Chlorophyll mutation frequency (\%)} = \frac{\text{Total number of chlorophyll mutants}}{\text{Total number of plants observed}} \times 100$$

$$\text{Morphological mutation frequency (\%)} = \frac{\text{Total number of morphological mutants}}{\text{Total number of plants observed}} \times 100$$

ANOVA and test of significance

The mean and coefficient of variations were calculated for all the quantitative traits from each treatment and replication. The calculated mean and CV were subjected to ANOVA (Analysis of variance) in RBD as indicated in Table 3. The test of significance difference was done by 'F' test. The critical difference of treatments were calculated to compare the mutagenic effects of the treatments.

Table 3. ANOVA for RBD

Source	d.f.	MS	'F' test
Replication	(r-1)	MS_r	MS_r/MS_e
Treatment	(t-1)	MS_t	MS_t/MS_e
Error	(t-1) (r-1)	MS_e	

Critical Difference (CD) = $(2.MS_e/r)^{\frac{1}{2}} \times t$ at (0.05) at error degree of freedom.

3.8.4 M₃ Generation

The selected M₂ progenies on the basis of yield were carried forward to M₃ generation. The mean yield of 10 randomly selected plants/progeny were recorded in each replication. This M₃ progeny yield/plant data were subjected to analysis in compact family block design as indicated in Table 4.

Table 4. ANOVA for compact family block design (Between treatment)

Source	d.f.	MS	'F' test
Replication (a)	(r-1)	MS _{r(a)}	MS _{r(a)} /MS _{e(a)}
Treatments	(g-1)	MS _g	MS _g /MS _{e(a)}
Error (a)	(g-1) (r-1)	MS _{e(a)}	

Table. 5. ANOVA for compact family block design (Between progenies within treatment)

Source	d.f.	Mean square within the treatment			
		1	2 12		
Replication (b)	(r-1)	MS _{r(b1)}	MS _{r(b2)} MS _{r(b12)}		
Progenies	(p-1)	MS _{p1}	MS _{p2} MS _{p12}		
Error (b)	(p-1) (r-1)	MS _{e(b1)}	MS _{e(b2)} MS _{e(b12)}		

Test of significance of difference among progenies of each treatment was calculated as

$$F = \frac{MS_{p1}}{MS_{e(b1)}} ; \frac{MS_{p2}}{MS_{e(b2)}} \dots\dots\dots$$

CD for treatment at (0.05) = $(2.MS_{e(a)}/r)^{\frac{1}{2}} \times t$ at (0.05) at error (a) d.f.

CD for progenies of each treatment = $(2 MS_{e(b1)}/r)^{\frac{1}{2}} \times t$ at (0.05) at error (b) d.f.

3.8.5 M₄ Generation

Estimation of variance components

The different variance components (genotypic, phenotypic, and environmental) were estimated for eight numbers of characters using the expected mean sum of squares of ANOVA table as indicated in Table 3.

Genotypic and Phenotypic variances were calculated following formula of Al - Jibouri et al. (1958).

$$\text{Genotypic variance } \delta_g^2 = \frac{MS(t) - MS_e}{r}$$

$$\text{Phenotypic variance } (\delta_p^2) = \delta_g^2 + \delta_e^2$$

Where, MS_t = Mean sum of square due to progeny

MS_e = Mean sum of square due to error
(which is equal to V_e)

r = Number of replications

δ_p = Phenotypic variance

δ_g^2 = Genotypic variance

δ_e^2 = Environmental variance = MS_e

The genotypic (δ_g^2) and phenotypic (δ_p^2) variances for different traits were used to calculate the genotypic (GCV) and phenotypic (PCV) coefficient of variation using the formula Burton (1952).

$$\text{Phenotypic coefficient of variation (PCV)} = \frac{\delta_p}{\bar{x}} \times 100$$

$$\text{Genotypic coefficient of variation (GCV)} = \frac{\delta_g}{\bar{x}} \times 100$$

where, δ_p and δ_g are the square root of phenotypic and

genotypic variance respectively and \bar{X} is the mean of the character.

Heritability

Heritability (broad sence) of the traits were estimated as the ratio of genotypic variance to levels of phenotypic variance following Hanson, et al. (1956) in the following manners.

$$h^2 \text{ heritability } (h^2) = \frac{\delta_g^2}{\delta_p^2} \times 100$$

Expected genetic advance

The genetic advance (GA) as expected from selections among mutatsns for different traits was calculated at 5 % selection intensity in accordance with Johnson et al. (1955).

$$GA = \delta_g \cdot k \cdot h.$$

where, δ_g = Square root of genotypic variance

h = Heritability coefficient

k = Standardized selection differential for specific selection intensity

(K = 2.06 at 5 % selection intensity)

GA was also expressed as percentage of the mean of the

$$\text{character, } GA \text{ (as \% of mean)} = \frac{GA}{\text{Mean}} \times 100$$

Estimation of covariance components

The ANCOVA between all possible pairs of characters was carried out on plot mean values. the mean sum of products from the ANCOVA was used to estimate the components of covariance as described in Table 6.

Table 6. ANCOVA with expectations of mean sum of products (MS_p) under model.

Source	d.f.	MS_p	Expected MS_p
Replication	(r-1)	MP_r	$\delta_{e(xy)}^2 + g \delta_{r(xy)}^2$
Cultures	(g-1)	MP_g	$\delta_{e(xy)}^2 + r \delta_{g(xy)}^2$
Error	(g-1) (r-1)	MP_e	$\delta_{e(xy)}^2$

The components of characters between two characters were estimated from the ANCOVA, using the expectation of MS_p (which are analogous to those of the mean squares in ANOVA) in the same way on variance components according to Al. Jibouri et al. (1958).

B = Genotypic correlation (r_g)

$$r_g = \frac{\delta_{g(xy)}}{\{\delta_{g(x)}^2 \cdot \delta_{g(y)}^2\}^{\frac{1}{2}}}$$

where, $\delta_{g(xy)}$ is the genotypic covariance between x and y
 $\delta_{g(x)}^2$ and $\delta_{g(y)}^2$ are the variance of character x and y.

Phenotypic correlation (r_p)

$$r_p = \frac{\delta_{p(xy)}}{\{\delta_{p(x)}^2 \cdot \delta_{p(y)}^2\}^{\frac{1}{2}}}$$

where, $\delta_{p(xy)}$ is the phenotypic covariance between x and y and
 $\delta_{p(x)}^2$, $\delta_{p(y)}^2$ are the variance of character x and y.

Test of significance.

Test of significance were done by t-test with (n-2) d.f.

$$t = \frac{r}{(1 - r^2)^{\frac{1}{2}}} (n-2)^{\frac{1}{2}}$$

where, r and n are the correlation coefficient and number of cultures respectively.

3.8.6 Multivariate approach in grouping the mutant lines

In view of the inadequacy of the single variable statistical approach, the data on different quantitative traits developed from the same parent were utilised to classify the mutant lines into different groups following multivariable approach of Mahalanobis D^2 Statistics following Rao (1952).

Multivariable test of significance

Utilising 'V' statistics, which in turn make use of Wilk's h criterion, (Rao 1948 and 1952), the multivariable test of significance, among the cultures with respect to multivariable correlated characters was performed.

If $|W|$ represent the error sum of square and the sum of product matrix,

$|B|$ the genotype sum of square and the sum of product matrix, then

$$\text{Wilk's criteria, } \lambda = \frac{|W|}{|B + N|} \quad \text{and}$$

$$\text{'V' Statistics} = - \left(m - \frac{p + q + 1}{2} \right) \log_e$$

p = Number of variable characters

q = Degrees of freedom of genotypes

m = Sum of degrees of freedom for error
and genotype variance.

This 'V' - statistics is distributed normally as χ^2 with $p \cdot q$ degrees of freedom and hence the required test of significance was performed using χ^2 distribution.

Analysis of divergence and grouping of mutant lines

Following Rao (1952) genetic divergence with respect to eight numbers of characters were estimated by Mahalanobis D^2 statistics. D^2 statistics, between any two mutant lines is given by the formula:

$$D^2_p = \frac{\sum_{i=1}^p \sum_{j=1}^p w^{ij} d_i d_j}{\sum_{i=1}^p \sum_{j=1}^p w^{ij} d_i d_j} \text{ where}$$

D^2_p = It is the inverse of W_{ij} , the common dispersion d_i and d_j are the differences in the mean of two populations for i^{th} and j^{th} characters.

As the method of analysis using this formula is complicated and labourious, the character means are transferred into sets of uncorrelated variables through pivotal condensation method of common dispersion matrix following Rao (1952). After transformation, the formula for genetic divergence was

$$D^2_p = \frac{\sum_{i=1}^p d_i^2}{\sum_{i=1}^p d_i^2} d_i^2$$

where, d_i is the difference between the transformed mean of any two populations for i^{th} character.

Statistical difference of significance between any two D^2 was tested by χ^2 with P degrees of freedom after computing all possible D^2_s among 47 mutant cultures of Dhauli and Nayagarh local. The assessment of relative contribution of individual characters to divergence was done by rank average method.

On the basis of D^2 value, the genotypes were grouped into clusters using Tocher's method as suggested by Rao (1952). The criterion used for grouping was that any two genotypes from the same cluster should have a smallest D^2 value than those from different clusters.

Canonical root method

Following Rao (1952) canonical analysis was also done and the Z_1 and Z_2 values corresponding to first two canonical vectors were plotted as a supplement to grouping by Tocher's method. This method is an extension of multiple regression analysis and is concerned with the study of association or interdependence of two sets of variables. This method can be used as a forecast model for clustering purpose as well. This canonical method of analysis involves calculations of canonical vectors or canonical roots and the two first canonical root values (Z_1 and Z_2) of each culture were taken for two dimensional presentation of the cultures in graph.

3.8.8 Path coefficient analysis

The path co-efficient analysis was carried out using the correlation co-efficient of seven characters as casual factors (independent variables) and yield as the effect (dependent variable). The relationship was obtained by solution of simultaneous equation which express the basic relationship between correlation and path coefficients as per the suggestion of De way and Lu (1959). The equations are as follows :

$$\begin{aligned} r_{1,y} &= P_{1,y} + r_{1,2}P_{2,y} + r_{1,3} P_{3,y} + \dots + r_{1,n} \cdot P_{ny}^T \\ r_{2,y} &= r_{2,1}P_{1,y} + P_{2,y} + r_{2,3} P_{3,y} + \dots + r_{2,n} \cdot P_{ny} \\ r_{n,y} &= r_{n,1} P_{1,y} + r_{n,2}P_{2,y} + r_{n,3} P_{3,y} + \dots + r_{n,n} \cdot P_{ny} \end{aligned}$$

where, r is the correlation coefficient between a pair of characters and P represents the direct effect of the character on yield. These simultaneous equations were solved in matrix method, where the direct effect matrix is obtained by multiplying the inverse of correlation matrix of these characters with the effected characters and then the indirect effects were calculated.

The path coefficient analysis was carried out at genotypic level, using the genotypic correlation coefficients of the mutant cultures.

3.8.9 Selection indices

Smith (1936) proposed a model for selection on several characters simultaneously using discriminant function of Fisher (1936). Since then the theory of selection model has been extended and modified in various ways by different authors to suit the requirement of practical breeding (Singh 1972).

Construction of selection indices

Genotypic and phenotypic variance among the characters provide the basis of constructing the selection indices. The selection indices are in this form.

$$I = b_1x_1 + b_x x_2 + \dots + b_n x_n$$

where, the X 's are the phenotypic values of the n characters included in the index construction; the b 's are the relative weight to be assigned to each characters for computing the index value and I is the index value of each entry on which selection is to be based. The ' b ' values are so estimated as to maximise expected genetic advance in the ultimate economic criterion from selection among the material.

In this present study, seed yield is considered as the ultimate economic criterion and thus the appropriate values of b's which would maximise expected genetic advance in yield from the index selection can be obtained by solution of the following simultaneous equations as shown by Smith (1936) and Hazel (1943).

$$b_1P_{1.1} + b_2P_{1.2} + \dots + b_nP_{1n} = G_{1.y}$$

$$b_1P_{2.1} + b_2P_{2.2} + \dots + b_nP_{2n} = G_{2.y}$$

$$b_1P_{n.1} + b_2P_{n.2} + \dots + b_nP_{n.n} = G_{n.y}$$

The (P) symbols in the earlier equations represent phenotypic variance/covariance among the characters and the (G) represent the genotypic covariance between any particular character and yield.

Genetic advance and relative efficiency

The expected genetic gain from selection based on the indices was calculated according to formula suggested by Smith (1936).

$$GA = k.(b_1G_{1y} + b_2G_{2y} + \dots + b_nG_{ny})^{\frac{1}{2}}$$

where, k = Standardised selection differential for specific selection intensity i.e. 1.40 at 20 % selection intensity.

Relative efficiency of selection based on the indices was assessed in terms of genetic advance from such selection relative to GA expected from direct selection on yield.

CHAPTER - III

MATERIALS AND METHODS

78.0 and 53.7 to 73.1 under different treatments of Dhauli and Nayagarh local respectively as against 96.0 and 98.0 respectively in controls. Germination in all the treated populations was reduced significantly in both the varieties as compared to their controls. Further, it was observed that combined treatments of mutagens affected germination more adversely than the single mutagenic treatment in both the varieties. Reduction in higher concentrations of single treatment was more than the lower concentrations. Different varieties responded differently to the mutagenic treatments. In variety Dhauli, NG treated populations recorded poor germination percentage followed by SA and EMS whereas, in Nayagarh local, population treated with SA were more affected following NG and EMS.

4.1.2 Survival

Survival (per cent) of the treated populations varied from 44.0 to 70.0 and 50.5 to 70.3 in Dhauli and Nayagarh local respectively as against 96.0 and 98.0 in the controls. All the treated populations of both the varieties recorded significant reduction in survival percentage as compared to their controls. Further, it was observed that with increase in dose of the mutagens, the survival percentage decreased considerably. The combine treatments of both the varieties, showed more reduction in survival than the single mutagenic treatment. In both the varieties, populations treated with NG showed lower survival percentage, followed by SA and EMS.

4.1.3 Seedling height

Seedling height (cm) was recorded 15 days after sowing in

the laboratory. The seedling height was reduced significantly in the treated populations of both the varieties from the control. The seedling height of the treated populations varied from 6.3 to 13.5 in Dhauli and 6.7 to 11.6 in Nayagarh local, while in the respective controls, it was 14.9 and 13.2. However, the reduction in height was dose dependant in other mutagenic treatments except NG. The seedling height was more reduced in the combine treatments than the single mutagenic treatment. In general, NG treatment was most effective in reducing seedling height followed by SA and EMS in both the varieties.

4.1.4 Pollen sterility

The pollen sterility (per cent) in the mutagenic treated populations was higher than the control in both the varieties (Tables 20 and 21). The pollen sterility was increased with increase in doses. The combined treatments of mutagen adversely affected the fertility as compared to the single treatment. The pollen sterility varied from 9.3 to 17.9 in the treated populations of Dhauli and 10.1 to 18.1 in Nayagarh local, as against 0.3 and 0.43 in the respective controls indicating differential response of varieties to mutagens. Populations treated with NG was more effective in reducing fertility in both the varieties. Population treated with SA and EMS responded differently to different varieties.

4.1.5 Seed sterility

The treated populations of both the varieties showed an increase in the seed sterility (per cent) as compared to control (Tables 20 and 21). The seed sterility varied from 7.5

Table 7. Effect of Single and Combine treatments of mutagens on different M₁ treated populations, Var : Dhauli.

Sl. No.	Treatment Code	Germination (%)	Germination as % of control	Survival (%)	Survival as % of control	Root Length (cm)	Shoot Length (cm)	Seedling height (cm)	Recovery Index (RI)	R.I as % of Control
1	E ₁	68.0*	70.83	66.0*	68.6	6.0*	7.5*	13.5*	891.0	61.4
2	E ₂	78.0*	81.12	70.0*	72.8	4.5*	5.6*	10.1*	707.0	49.4
3	S ₁	64.0*	66.56	60.0*	62.4	4.2*	5.6*	10.1*	552.0	38.1
4	S ₂	72.0*	74.88	66.0*	68.6	4.0*	4.9*	8.9*	587.4	35.2
5	N ₁	60.0*	62.40	58.0*	60.3	3.7*	4.3*	8.0*	464.0	32.0
6	N ₂	64.0*	66.56	66.0*	68.6	3.8*	4.5*	8.3*	547.8	37.4
7	E ₂ S ₂	68.0*	70.83	56.0*	58.2	3.1*	3.9*	7.0*	448.0	30.9
8	E ₂ N ₂	58.0*	60.32	50.0*	52.0	3.3*	4.7*	8.0*	400.0	27.6
9	S ₂ N ₂	54.0*	56.16	50.0*	52.0	3.5*	4.6*	8.1*	405.0	27.9
10	E ₁ S ₁	68.0*	70.72	60.0*	62.4	3.7*	4.4*	8.1*	405.0	27.9
11	E ₁ N ₁	66.0*	68.64	64.0*	66.6	3.3*	4.3*	7.6*	436.4	30.5
12	S ₁ N ₁	67.0*	69.68	66.0*	68.4	3.1*	4.3*	7.4*	488.4	34.2
13	E ₂ S ₁	68.0*	70.83	60.0*	62.4	3.7*	4.8*	8.5*	510.0	35.7
14	E ₂ N ₁	66.0*	68.64	54.0*	38.2	2.8*	3.5*	6.3*	340.6	23.8
15	S ₂ N ₁	66.0*	68.64	50.0*	50.0	3.0*	3.7*	6.7*	335.0	23.4
16	E ₁ S ₂	60.0*	62.40	56.0*	58.2	3.6*	4.1*	7.7*	431.2	30.2
17	E ₁ N ₂	56.0*	58.24	52.0*	54.0	3.8*	4.7*	8.5*	442.0	30.9
18	S ₁ N ₂	52.0*	54.08	44.0*	45.8	4.0*	5.7*	9.7*	426.8	29.9
19	Control	96.0	100.00	96.0	100.0	6.7	8.2	14.9	1430.4	100.0
C.D. (0.05)		8.8	---	4.6	---	0.5	0.7	0.8	---	---

* Significant decrease at P = 0.05

Table 8. Effect of Single and Combine treatments of mutagens on different M₁ treated populations, Var : Nayagarh local.

Sl. No.	Treatment Code	Germination (%)	Germination as % of control	Survival (%)	Survival as % of control	Root Length (cm)	Shoot Length (cm)	Seedling height (cm)	Recovery Index (RI)	R.I as % of Control
1	E ₁	65.3*	66.0	60.0*	61.2	5.6	6.0*	11.6*	696.0	53.6
2	E ₂	73.1*	74.5	70.3*	71.7	4.3*	5.7*	10.0*	703.0	54.1
3	S ₁	62.7*	63.9	56.3*	57.1	4.4*	5.8*	10.2*	571.2	44.0
4	S ₂	69.3*	71.2	70.0*	71.4	4.2*	5.3*	9.5*	665.0	51.2
5	N ₁	65.7*	60.9	57.0*	58.1	3.8*	4.0*	7.8*	446.6	34.4
6	N ₂	68.8*	64.1	61.7*	62.9	3.6*	4.7*	8.3*	512.1	39.4
7	E ₂ S ₂	60.3*	61.5	56.3*	57.4	3.0*	4.1*	7.1*	399.7	30.8
8	E ₂ N ₂	53.7*	54.8	50.5*	51.5	3.5*	3.9*	7.4*	373.7	28.8
9	S ₂ N ₂	58.2*	59.4	53.4*	54.4	3.8*	3.9*	7.7*	411.2	31.7
10	E ₁ S ₁	65.3*	66.6	53.3*	54.4	3.8*	4.2*	8.0*	435.0	33.5
11	E ₁ N ₁	66.1*	67.4	62.5*	63.7	3.5*	4.1*	7.6*	475.0	36.6
12	S ₁ N ₁	65.3*	66.6	57.3*	58.4	3.0*	3.7*	6.7*	389.9	29.6
13	E ₂ S ₁	66.1*	67.4	60.0*	61.2	4.0*	4.3*	8.3*	498.0	38.3
14	E ₂ N ₁	62.7*	63.9	58.3*	59.5	4.3*	4.8*	9.1*	530.5	40.8
15	S ₂ N ₁	60.3*	61.5	51.2*	52.2	3.5*	4.1*	7.6*	389.1	29.9
16	E ₁ S ₂	58.3*	59.4	51.8*	52.3	4.3*	4.9*	9.2*	476.6	36.7
17	E ₁ N ₂	58.7*	59.9	52.3*	53.3	4.0*	4.3*	8.3*	434.1	33.4
18	S ₁ N ₂	62.8*	64.0	56.3*	57.4	3.7*	4.5*	8.2*	461.7	35.3
19	Control	98.0	100.0	98.0	100.0	6.0	7.2	13.2	1293.6	100.0
C.D. (0.05)		1.1	---	2.4	---	0.7	0.9	1.5	---	---

* Significant decrease at P = 0.05

to 12.5 and 8.1 to 14.2 in the treated populations of Dhauri and Nayagarh local respectively as against 0.7 and 1.0 in the controls. The seed sterility was observed to be dose dependent. The combine mutagenic treated populations showed further decrease in seed fertility compared to single mutagenic treatment. In general, NG treated populations recorded higher seedling sterility followed by SA and EMS in both the varieties.

4.1.6 Root and shoot length

The root and shoot length (cm) of the treated populations of both the varieties was significantly reduced as compared to respective controls (Tables 7 and 8). In the treated populations of Dhauri, the root and shoot length varied from 2.8 to 6.0 and 3.5 to 7.5 respectively as against 6.7 and 8.2 in control. The root and shoot length in the treated populations of Nayagarh local varied from 3.0 to 5.6 and 3.7 to 6.0 whereas it was 6.0 and 7.2 in the control respectively.

The root and shoot length was reduced in the treated populations of both the varieties than the control. In both the varieties, these characters were adversely affected in the combine treated populations than the single mutagenic treatment. Populations treated with NG exhibited more reduction in root and shoot length in both the varieties followed by SA and EMS. The root length of the treated populations, in both the varieties, was more affected than the shoot length.

4.1.7 Plant height

The plant height (cm) was recorded at maturity. The E_1N_1 , S_1N_1 , E_1S_2 and S_1N_2 treated populations of Dhauri showed

significant reduction in plant height as compared to control (Tables 9 and 10). All the treated populations of Nayagarh local except S_1 and S_1N_2 exhibited significant reduction in plant height from control. The plant height in the treated populations of Dhauri and Nayagarh local varied from 18.3 to 30.9 and 17.4 to 28.0 respectively. The height of the control plant was 28.8 and 27.7 in Dhauri and Nayagarh local respectively. Populations treated with EMS showed more reduction in height followed by SA and NG in Dhauri. The height was more reduced in NG treated population followed by EMS and SA in Nayagarh local. The combine treatment of mutagens responded differently as compared to the single treatment in both the varieties.

4.1.8 Pods per plant

The number of pods per plant in the treated populations of Dhauri and Nayagarh local varied from 8.40 to 16.8 and 8.8 to 17.7 respectively as against 15.5 and 16.9 in the respective controls. All the mutagenic treated populations of Dhauri except E_1 , S_2 , E_2N_1 , S_2N_1 and S_1N_2 showed significant reduction in pods per plant than the control. There was significant reduction in pods per plant in all the treated population of Nayagarh local except E_2 , S_2 and S_1N_2 mutagenic treated populations than the control.

4.1.9 Seeds per pod

The seeds per pod in the populations treated with the different mutagens varied from 2.91 to 5.5 in Dhauri and from 2.8 to 4.5 in Nayagarh local. The control plant of Dhauri and Nayagarh local produced 4.7 and 4.6 number of seeds per pod

respectively. The populations treated with S_2 , N_2 , E_2S_2 , E_2N_2 , S_2N_2 , S_1N_1 , E_1N_1 and E_1S_2 showed a significant reduction in seeds per pod than control, while, E_2S_1 recorded significant increase in Dhauri. In Nayagarh local, except S_2N_1 , all other treated populations observed significant lower seeds per pod than control. The two varieties responded differently to different mutagens with respect to induction of mutation in this character.

4.1.10 100-seed weight

The variation noticed in 100-seed weight (g) in the treated populations was from 2.10 to 3.70 in Dhauri and from 1.70 to 3.40 in Nayagarh local indicating differential response of varieties to different mutagens. The 100-seed weight of 3.90 and 3.80 was recorded in the control plant of Dhauri and Nayagarh local respectively. In Dhauri, except populations treated with E_2N_2 and E_1S_1 , other treated populations observed a significant reduction in 100-seed weight than control. The observed 100-seed weight was significantly lower in all the treated population of Nayagarh local as compared to control.

4.1.11 Plant yield

The variation in yield/plant (g) of Dhauri ranged from 1.20 to 4.50 as against 3.3 in control. All the treated populations except S_2 and E_1S_1 recorded significant reduction in plant yield than the control. The S_2 treated population observed significant increase in plant yield than control. In Nayagarh local the variation observed in plant yield was 1.3 to 3.8 as against 3.3 of the control. All the treated populations except E_1 , E_2 , S_1 , S_2 , E_1S_1 and S_1N_2 observed significant

reduction in plant yield. The decrease in plant yield in the combine treated populations was more than the single mutagenic treatment.

4.1.12 Days to maturity

The period taken for maturity (days) is indicated in Tables 9 and 10. The maturity of the treated populations of Dhauli and Nayagarh local ranged from 57 to 65 and 62 to 70 respectively as against 62 and 68 in the controls. In Dhauli, the populations treated with E_2N_2 , S_2N_2 , E_1S_1 , E_2S_1 , E_2N_2 , S_2N_1 and E_1S_1 recorded significant reduction in maturity duration than the control. In case of Nayagarh local, populations treated with S_1 , S_2 , N_2 , E_2S_2 and E_2N_1 showed significant reduction in days to maturity than the control.

4.1.13 Recovery Index

The Recovery Index (RI) of the single and combine treatments of mutagens in M_1 generation was calculated by using the formula $\bar{X} \times S$; where, \bar{X} is the mean of seedling height and 'S' is the corresponding percentage of survival in the same treatment. It was observed that the RI of the combine treatment of mutagens was lower than the single treatment (Tables 7 and 8). In both the varieties lower R.I. was observed in treated populations than the control.

The coefficient of interaction 'K' was less than one indicating the additive type of action in all the combine treated populations of Dhauli. The 'K' value ranged from 0.66 to 0.94 in Dhauli (Table 13). In case of Nayagarh local, this coefficient of interaction ranged from 0.77 to 1.46. In the combine treated populations of Nayagarh local 'K' was also less

Table 9. Mean, Range of M₁ quantitative characters of different mutagenic treated populations, Var - Dhauili.

Sl. No.	Treatment Code	Plant Height (cm)			Pods / Plant			Seeds / Pod			100-Seed Weight (g)			Single Plant yield (g)			Maturity (days)		
		Range	Mean	Percent of Control	Range	Mean	Percent of Control	Range	Mean	Percent of Control	Range	Mean	Percent of Control	Range	Mean	Percent of Control	Range	Mean	Percent of Control
1	E ₁	20-27	25.5	88.5	11-17	14.8	94.5	4-7	5.0	106.5	2.0-3.3	2.4*	61.5	4.0-4.9	4.0**	121.2	55-70	63	101.6
2	E ₂	23-30	27.0	93.7	8-16	10.6*	68.4	4-7	4.7	100.0	2.8-4.3	3.2*	82.0	3.7-5.0	4.1**	124.2	58-67	62	100
3	S ₁	23-31	26.3	91.3	8-16	10.1*	65.2	4-6	4.2	89.4	2.7-3.4	3.0*	76.9	2.3-3.7	2.8*	84.8	60-71	65**	104.8
4	S ₂	21-34	29.3	101.7	11-15	14.1	90.0	3-4	4.0*	85.1	2.0-3.2	2.4*	61.5	4.0-5.0	4.5**	136.4	60-72	64	103.2
5	N ₁	26-33	30.9	107.3	7-14	8.4*	54.2	3-4	4.3	91.5	2.0-2.8	2.2*	56.4	1.9-3.0	2.3*	69.7	55-68	63	101.6
6	N ₂	24-32	27.4	95.1	9-13	10.9*	70.3	3-4	3.1*	65.9	1.7-2.5	2.0*	51.3	1.7-3.3	2.0*	90.9	54-67	62	100
7	E ₂ S ₂	23-32	27.7	96.2	8-15	10.3*	70.3	3-5	4.0*	85.1	1.7-3.0	2.1*	53.8	1.5-3.0	1.8*	54.5	54-68	60	96.8
8	E ₂ N ₂	25-30	28.4	98.6	7-14	9.4*	60.4	2-4	2.9*	61.7	2.8-4.1	3.3	84.6	1.6-3.8	2.1*	63.3	50-64	59*	95.2
9	S ₂ N ₂	22-31	26.3	91.3	7-13	8.8*	56.8	4-5	4.0*	85.1	1.7-2.3	2.3*	58.9	1.8-2.7	2.1*	63.3	50-63	58*	93.5
10	E ₁ S ₁	25-31	27.6	95.8	9-17	11.4*	73.7	4-6	5.0	106.5	3.0-4.4	3.7	94.9	3.2-4.7	3.6	109.1	51-69	59*	95.2
11	E ₁ N ₁	17-26	20.6*	71.5	7-15	8.6*	55.5	2-4	2.9*	51.1	1.7-2.5	2.3*	58.9	1.8-2.7	2.0*	60.6	50-65	60	96.8
12	S ₁ N ₁	15-23	18.6*	64.6	8-12	10.6*	68.4	3-4	3.7*	78.7	2.0-3.4	2.6*	66.6	1.0-1.5	1.2*	36.4	52-68	61	98.4
13	E ₂ S ₁	24-33	27.4	95.1	9-15	11.5*	74.2	3-5	5.5**	74.5	2.1-2.9	2.7*	69.2	1.2-2.0	1.6*	48.5	52-65	59*	95.2
14	E ₂ N ₁	24-33	27.4	95.1	11-20	14.3	92.2	4-6	5.2	110.6	2.8-3.0	3.0*	76.9	2.5-3.0	2.7*	81.8	50-63	57*	91.9
15	S ₂ N ₁	19-31	24.0	83.3	14-22	16.8	108.4	4-5	4.2	89.4	2.0-3.7	2.4*	61.5	1.8-2.6	2.2*	66.6	51-65	58*	93.5
16	E ₁ S ₂	19-27	22.5*	78.1	9-16	10.2*	65.8	3-4	3.0*	63.8	2.0-2.9	2.4*	61.5	1.8-2.5	2.0*	90.9	52-70	59*	95.2
17	E ₁ N ₂	20-27	23.1	80.2	8-15	9.7*	62.2	4-5	4.1	87.2	1.8-2.5	2.1*	53.8	2.0-3.4	2.7*	81.8	55-70	61	98.4
18	S ₁ N ₂	16-33	20.7*	71.9	12-19	14.5	93.5	5-6	5.1	108.5	2.4-3.1	2.9*	74.3	1.4-2.1	1.7*	51.5	54-68	60	96.8
19	Control	26-30	28.8	100.0	15-17	15.5	100.0	4-5	4.7	100.0	3.6-3.9	3.9	100.0	3.3-3.6	3.3	100	60-63	62	100
C.D. (0.05)			5.9			2.7			0.6			0.6			0.4				2.4

* Significant decrease at P = 0.05

** Significant increase at P = 0.05

SI No.	Treatment Code	Plant Height (cm)			Pods / Plant			Seeds / Pod			100-Seed Weight (g)			Single Plant yield (g)			Maturity (days)		
		Range	Mean	Percent of Control	Range	Mean	Percent of Control	Range	Mean	Percent of Control	Range	Mean	Percent of Control	Range	Mean	Percent of Control	Range	Mean	Percent of Control
1	E ₁	20-28	24.6*	88.8	11-20	14.5*	86.1	3.0-7.5	4.0*	86.9	2.9-4.1	3.1*	77.2	3.2-4.2	3.5	107.0	60-75	70	102.9
2	E ₂	16-27	20.2*	72.9	14-21	17.7	104.7	3.1-7.1	4.0*	86.9	2.7-4.3	2.9*	74.7	3.0-4.0	3.0	91.7	60-73	68	100.0
3	S ₁	20-29	28.0	101.1	9-15	10.7*	63.7	3.0-4.7	3.9*	84.8	2.6-4.7	2.9*	73.7	3.2-3.9	3.3	91.7	58-78	64*	94.1
4	S ₂	16-28	21.7*	78.3	12-21	16.7	90.6	2.2-4.8	3.0*	56.5	2.6-4.0	2.7*	68.6	3.1-4.0	3.8	115.6	51-65	62*	91.2
5	N ₁	20-28	23.8*	85.9	7-10	8.8*	52.0	3.0-5.8	3.5*	76.1	1.0-2.7	1.9*	48.6	2.3-3.1	2.8*	86.8	53-70	66	97.0
6	N ₂	16-25	19.4*	70.0	7-18	9.9*	58.6	2.7-5.8	3.3*	71.7	2.5-4.1	3.0*	75.9	2.2-3.4	2.9*	87.5	60-70	64*	94.1
7	E ₂ S ₂	20-30	25.4*	91.7	7-14	10.3*	60.9	2.8-7.2	3.6*	78.3	1.7-3.1	2.1*	52.1	1.9-2.7	2.3*	68.5	58-69	64*	94.1
8	E ₂ N ₂	16-29	22.3*	80.5	5-11	8.8*	57.1	2.9-5.6	3.4*	78.9	2.0-3.1	2.9*	73.9	2.4-2.9	2.6*	74.6	57-67	65	95.6
9	S ₂ N ₂	24-30	27.3	98.6	7-14	10.3*	60.9	3.0-5.7	3.7*	80.4	1.9-3.7	2.1*	54.2	1.0-2.9	1.5*	45.9	60-72	66	97.0
10	E ₁ S ₁	14-25	18.9*	68.2	13-19	15.8*	93.8	2.3-6.2	3.8*	60.9	1.0-4.1	1.9*	49.4	3.0-3.8	3.1	95.1	58-71	67	98.5
11	E ₁ N ₁	18-25	21.9*	79.1	7-14	15.8*	68.0	2.9-4.3	3.0*	65.2	1.3-3.1	1.9*	49.1	1.0-1.8	1.3*	38.5	57-70	67	98.5
12	S ₁ N ₁	17-22	19.6*	70.7	7-15	11.5*	59.5	2.4-5.2	2.8*	60.9	2.0-3.1	2.2*	55.2	1.2-1.9	1.7*	51.1	60-70	68	100.0
13	E ₂ S ₁	24-33	28.6	103.2	7-14	10.0*	63.8	2.8-5.4	3.0*	65.2	1.6-3.0	1.7*	44.0	1.0-1.6	1.4*	40.2	60-71	65	95.6
14	E ₂ N ₁	14-21	17.4*	62.8	9-15	10.8*	74.6	3.0-5.8	3.8*	82.6	2.5-3.5	2.6*	64.8	2.1-3.2	2.4*	74.6	58-68	64*	94.1
15	S ₂ N ₁	19-24	22.0*	79.4	10-20	12.6*	84.0	3.7-7.2	4.5	97.8	2.3-3.4	2.8*	71.4	1.7-2.9	2.0*	61.2	60-71	65	95.6
16	E ₁ S ₂	16-31	20.4*	73.6	7-15	14.2*	63.4	2.1-4.2	3.0*	65.2	2.0-2.8	2.7*	67.3	1.0-2.2	1.6*	48.0	62-71	67	98.5
17	E ₁ N ₂	19-24	20.6*	74.4	9-13	10.7*	63.3	3.0-6.8	4.0*	86.9	2.1-3.1	2.7*	68.9	1.2-2.4	1.8*	54.1	61-73	65	95.6
18	S ₁ N ₂	13-25	18.9*	68.2	15-21	16.5	97.6	3.5-7.2	4.5	97.8	2.8-3.7	3.4*	85.1	2.8-4.0	3.5	107.5	62-71	67	98.5
19	Control	25-29	27.7	100.0	14-17	16.9	100.0	4.0-5.0	4.6	100.0	3.7-4.0	3.8	100.0	3.1-3.5	3.3	100.0	67-70	68	100.0
C.D. (0.05)		---	1.7	---	0.8	---	0.3	---	0.3	---	0.3	---	0.3	---	0.3	---	---	---	3.5

* Significant decrease at P = 0.05

** Significant increase at P = 0.05

than one except E_2S_2 , S_2N_2 , E_1S_1 , E_1N_1 and S_1N_2 .

4.2 M₂ GENERATION

A wide range of variability affecting different vegetative and yield components by mutagenic treatments was observed in M₂ in this investigation. The frequency and spectrum of chlorophyll and morphological mutations observed in M₂ generation is presented in the following sections.

4.2.1 Chlorophyll mutants

The chlorophyll mutants were scored daily when the seedlings were 5 to 15 days old. To estimate chlorophyll mutation 500 and 700 plants were observed daily in each treatment and the result on frequency and spectrum of chlorophyll mutation is presented in Tables 11 and 12.

The chlorophyll mutation (per cent) of the treated populations varied from 1.39 to 3.11 in Dhauli and 1.04 to 3.44 in Nayagarh local. There was no evidence of chlorophyll mutations in both the controls. The S_2N_2 treated populations of Dhauli and Nayagarh local recorded the highest chlorophyll mutation frequency. Populations treated with S_1 in both the varieties recorded the lowest chlorophyll mutations frequency of 1.39 and 1.04 per cent respectively. The chlorophyll mutation frequency recorded in the combine treated populations were higher than single mutagenic treatment in both the varieties.

Spectrum of different chlorophyll mutations observed in M₂ generation were albina, xantha, chlorina, viridis and sectorial (Tables 11 and 12). In Dhauli, chlorina type mutants were observed in maximum following followed by xantha, albina,

viridis and sectorial in descending order. In Nayagarh local, chlorina, xantha, viridis, albina and sectorial type mutants were observed in descending order. In both the varieties, NG treated populations recorded high mutation frequency.

4.2.2 Morphological mutations

The morphological mutants included those affecting morphology of different plant parts, such as, leaf size and shape, plant height, pod size, pod colour, seed size and colour, stem colour, plant type, etc.

The frequency of morphological mutations (per cent) ranged from 4.10 to 7.99 in Dhauli and 2.90 to 8.18 in Nayagarh local. In Dhauli, populations treated with SA showed maximum morphological mutations followed by EMS and NG. Treatment with NG followed by EMS and SA in descending order, produced more morphological mutations in Nayagarh local. However, highest mutations frequency was observed in S_2 and N_2 treated populations of Dhauli and Nayagarh local respectively. The lowest mutations frequency of 4.10 and 2.90 was observed in S_1N_1 and E_2N_2 treated populations of Dhauli and Nayagarh local respectively.

4.2.3 Total macromutations

The total macromutations included both chlorophyll and morphological mutations. The total mutation frequency (per cent) varied from 6.33 to 9.76 in Dhauli and 5.64 to 10.00 in Nayagarh local as against no occurrence of macromutations in controls. The total mutation frequency was decreased in the combine treated populations of both the varieties, as compared to single mutagenic treated populations except E_1N_2 of Dhauli.

Table 11. Spectrum of different types of mutants (Chlorophyll morphological and total). M₂ generation, Var - Dhauli.

Mutagenic Treatment

No. of plants observed	E ₁	E ₂	S ₁	S ₂	N ₁	N ₂	E ₂ S ₂	E ₂ N ₂	S ₂ N ₂	E ₁ S ₁	E ₁ N ₁	S ₁ N ₁	E ₂ S ₁	E ₂ N ₁	S ₂ N ₁	E ₁ S ₂	E ₁ N ₂	S ₁ N ₂	C	Total	(%)
	541	545	572	563	585	650	563	620	610	573	582	585	631	682	651	617	485	687	-	10,832	

Chlorophyll mutations

Chlorina	2	3	3	4	2	3	5	4	5	5	3	5	4	5	4	7	3	5	---	72	0.66
Xantha	2	3	2	3	2	3	6	3	4	4	5	4	4	4	2	4	4	5	---	64	0.59
Albina	2	---	2	2	2	3	3	3	4	2	4	3	3	3	3	3	2	4	---	48	0.44
Viridis	1	1	---	1	3	2	2	4	3	3	2	2	2	3	2	---	1	3	---	35	0.32
Sectorial	1	2	1	---	1	2	---	3	3	1	1	3	1	1	4	2	3	3	---	32	0.29
Total	8	9	8	10	10	13	16	17	19	15	15	17	14	16	15	16	13	20	---	251	2.31
Frequency (%)	1.47	1.65	1.39	1.77	1.71	2.0	2.84	2.74	3.11	2.61	2.58	2.90	2.21	2.34	2.30	2.59	2.68	2.91	---	2.31	---

Morphological Mutants

Tall	6	10	5	6	5	2	5	6	4	3	3	2	3	5	10	4	6	5	---	---	---
Dwarf	5	4	5	4	7	8	5	7	7	10	5	3	4	2	7	3	3	4	---	---	---
Trailing	1	---	---	1	1	---	1	2	---	2	---	1	2	3	1	2	3	1	---	---	---
Early	7	5	5	5	5	7	3	5	7	4	4	4	2	2	1	1	---	4	---	---	---
Late	6	6	4	6	3	7	5	5	2	---	---	3	3	3	6	4	8	5	---	---	---
Quardifoliate	---	3	2	4	7	3	5	1	1	---	---	3	4	---	---	---	---	3	---	---	---
Pontafoliate	5	---	5	7	2	3	1	2	4	2	2	2	3	6	3	5	3	3	---	---	---
Profused branched	---	---	3	1	1	1	7	---	---	7	3	---	1	---	---	4	3	2	---	---	---
More poded	4	---	---	5	---	4	1	---	5	2	2	2	1	2	3	5	4	4	---	---	---
Bushy	1	2	1	3	4	2	---	---	2	---	3	2	2	4	3	3	3	2	---	---	---
Stunted plant	---	1	---	2	2	---	---	---	---	2	1	2	1	---	---	---	---	4	---	---	---
Erect	1	3	---	1	1	---	1	---	1	---	1	---	---	---	1	1	---	1	---	---	---
Total	36	34	30	45	38	37	34	28	33	32	24	24	26	28	35	32	33	38	---	---	---
Frequency (%)	6.65	6.23	5.24	7.99	6.49	5.69	6.03	4.51	5.40	5.58	4.12	4.10	4.12	4.10	5.37	5.18	6.80	5.53	---	---	---
Total mutation	44	43	38	55	48	50	50	45	52	47	39	41	40	44	50	48	46	58	---	---	---
Frequency (%)	8.12	7.88	6.63	9.76	8.20	7.69	8.87	7.25	8.51	8.46	6.70	7.00	6.33	6.44	7.67	7.77	9.48	8.44	---	---	---

Table 12. Spectrum of different types of mutants (Chlorophyll morphological and total). M₂ generation, Var - Nayagarh local.

No. of plants observed	Mutagenic Treatment														Total	C	S ₁ N ₂	E ₁ N ₂	E ₁ S ₂	S ₂ N ₁	E ₂ N ₁	E ₂ S ₁	S ₁ N ₁	E ₁ N ₁	E ₁ S ₁	S ₂ N ₂	E ₂ N ₂	E ₂ S ₂	N ₂	N ₁	S ₂	S ₁	E ₂	E ₁	Total	(%)
	620	610	673	595	585	531	651	517	585	595	673	610	620	563																						
Chlorophyll mutations																																				
Chlorina	4	3	2	4	4	6	4	4	4	2	2	5	6	4	5	4	4	5	5	4	4	5	5	5	5	71	0.63									
Xantha	1	3	2	2	4	3	6	5	3	3	4	4	4	3	3	4	4	3	3	3	3	3	3	3	3	61	0.54									
Viridis	1	---	1	1	4	4	6	3	4	3	4	4	2	3	2	4	4	3	2	3	3	2	4	4	46	0.41										
Albina	1	2	1	2	4	4	3	5	3	3	4	4	2	---	---	---	---	---	---	---	---	---	---	---	42	0.37										
Sectorial	---	1	1	---	3	4	4	2	3	2	1	3	3	4	2	2	4	2	2	4	2	2	2	37	0.32											
Total	7	9	7	9	17	21	23	19	17	13	18	17	17	14	12	19	14	12	19	14	12	19	19	257	---	---										
Frequency (%)	1.29	1.51	1.04	1.17	3.01	2.74	3.44	3.41	3.19	2.90	2.76	3.28	2.39	2.41	3.15	2.41	2.44	2.76	3.28	2.39	2.41	3.15	2.28	2.28	---	---										
Morphological Mutants																																				
Tall	6	12	4	7	9	5	2	5	3	2	4	5	4	3	5	4	2	4	4	3	5	4	3	5	---	---										
Dwarf	7	7	6	8	12	7	8	8	8	5	7	3	3	5	4	7	4	4	4	5	4	4	4	7	---	---										
Trailing	---	1	2	2	---	---	4	3	3	---	---	2	2	---	---	2	---	---	---	---	---	---	---	---	---	---										
Early	7	2	9	6	3	5	5	2	2	5	5	4	3	3	3	5	5	4	3	3	3	3	5	---	---											
Late	5	4	4	3	5	3	4	1	4	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---										
Quardifoliate	---	3	2	5	2	8	---	4	4	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---										
Pontafoliate	---	3	4	1	2	5	---	3	3	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---										
Profused branched	3	---	---	4	---	---	4	1	4	2	4	4	---	---	---	---	---	---	---	---	---	---	---	---	---	---										
More poded	2	3	3	4	---	2	4	---	4	4	2	3	3	2	2	2	4	2	3	2	2	2	2	---	---											
Bushy	4	3	1	2	---	2	4	1	1	4	4	5	4	4	4	2	4	4	5	4	4	4	2	---	---											
Stunted plant	1	2	2	1	---	1	1	1	1	---	---	2	---	---	---	---	---	---	---	---	---	---	---	---	---	---										
Erect	1	---	---	1	1	1	---	1	1	1	1	---	---	1	---	---	---	---	---	1	---	---	---	---	---	---										
Total	36	40	37	44	34	36	39	28	23	23	37	24	23	25	40	23	23	37	24	23	25	40	40	---	---											
Frequency (%)	6.65	6.72	5.50	5.76	6.03	2.90	5.90	5.79	4.70	3.93	5.68	4.64	3.93	5.04	6.64	3.93	4.33	5.68	4.64	3.93	5.04	6.64	6.64	---	---											
Total mutation	43	49	44	53	51	35	57	62	47	40	36	55	41	37	59	37	37	55	41	37	37	59	59	---	---											
Frequency (%)	7.94	8.23	6.54	6.93	9.04	5.64	9.34	9.20	7.89	6.83	6.77	8.44	7.92	6.32	7.45	6.32	6.77	8.44	7.92	6.32	7.45	9.80	9.80	---	---											

The SA treated populations of Dhauli recorded highest total mutation frequency followed by EMS and NG. In case of Nayagarh local, NG treated population followed by EMS and SA recorded highest total mutation frequency in descending order.

4.3 INDUCED CHANGE IN MEAN OF THE QUANTITATIVE CHARACTERS

Mutation in quantitative characters can be best assessed by estimation of mean and variance of the characters controlled by polygenes each with small effect. Induction of micromutations on quantitative characters would result in an increase in variability with or without affecting mean. Hence, these two parameters were estimated for all the eight characters of the two varieties and are presented in Tables 14, 15, 16 and 17.

4.3.1 Plant height

The plant height (cm) of the treated populations of Dhauli and Nayagarh local varied from 38.25 to 51.10 and 26.30 to 42.60 respectively. The average plant height of Dhauli and Nayagarh local was 46.10 and 42.60 respectively under normal condition. Compared to control, significant increase in plant height was obtained in the treated populations of E_2S_2 , E_2N_2 , E_2N_1 of Dhauli. There was significant decrease in plant height in the treated populations of S_1 , S_2 , N_1 , N_2 , E_1N_1 , S_2N_1 , E_1S_2 , E_1N_2 and S_1N_2 of the same variety. All the treated populations of Nayagarh local showed a decrease in plant height as compared to control. However, negative shift of mean in Nayagarh local was significant only in the treated populations of E_1 , E_2 , S_1 , S_2 , E_2S_2 , E_1S_1 , E_1N_1 , S_1N_1 , E_2S_1 , E_1N_2 and S_1N_2 . In Dhauli, in the treated populations of NG, the plant height was most

adversely affected followed by SA and EMS. Reduction in plant height was highest in the populations of Nayagarh local treated with EMS followed by SA and NG.

4.3.2 Pods per plant

Average number of pods per plant in the treated populations of Dhauli and Nayagarh local varied from 15.90 to 29.70 and 11.30 to 21.60 respectively, as against 18.80 and 16.50 in the respective controls. In both the varieties, the average pods per plant of the treated populations varied both on positive and negative directions of the control. There was a significant increase in pods per plant in the treated populations of E_2 , S_2 , E_1S_1 , E_2N_1 , S_2N_1 and S_1N_2 of Dhauli and E_2 , N_1 , N_2 , E_2S_2 , E_1S_1 , S_1N_2 of Nayagarh local. The treated populations of EMS recorded highest pod number followed by SA and NG in Dhauli. In Nayagarh local NG treated populations followed by EMS and SA showed maximum number of pods per plant.

4.3.3 Pod length

The pod length (cm) in the treated populations varied from 6.20 to 9.30 in Dhauli and 5.20 to 7.90 in Nayagarh local. The mean pod length observed in control plants was 6.40 and 6.50 in Dhauli and Nayagarh local respectively. The treated populations showed both positive and negative shift in the mean of the character over control in both the varieties. Treated populations of E_1 , S_2 , E_1S_1 , E_2S_1 and S_1N_1 showed significant increase in pod length over control in Dhauli. There was a significant increase in pod length only in the treated populations N_2 of Nayagarh local. On an average the pod length was highest in populations treated with EMS followed by SA and

NG in Dhauli. While in Nayagarh local this was highest in NG treated populations followed by SA and EMS.

4.3.4 Seeds per pod

The seeds per pod varied from 3.50 to 5.90 and 3.10 to 5.60 respectively in the treated populations of Dhauli and Nayagarh local as against 4.60 and 4.20 in their respective controls. In Dhauli treated populations of E_1 , E_2 , S_1 , S_2 , N_1 , E_1S_1 , E_2N_1 , S_2N_1 and S_1N_2 recorded a significant increase in the seeds per pod over control. Treated populations of E_1 , E_2 , S_2N_1 and S_1N_2 of Nayagarh local also showed a significant increase in seeds per pod over control. In both the varieties in the treated populations of EMS recorded highest number of seeds per pod as far as single mutagenic treatment was concerned. However, in the treated populations of S_1N_2 produced highest seeds/pod, among all the treatments of both the varieties.

4.3.5 100-seed weight

The average 100-seed weight (g) in the treated populations varied from 2.70 to 4.58 in Dhauli and 2.33 to 4.42 in Nayagarh local. The same in the control populations of Dhauli and Nayagarh local was 3.40 and 3.84 respectively. Treated populations of E_1 , E_2 , S_1 , S_2 , E_2N_2 , E_1S_1 , E_2N_1 , S_2N_1 and S_1N_2 in Dhauli and S_1 , E_2N_2 and S_1N_2 in Nayagarh local showed significant increase in 100-seed weight as compared to control. However, negative and positive shift in mean in 100-seed weight was observed in the other treated populations of both the varieties.

Table 14. Mean values of quantitative traits in M₂ generation of different mutagen treated populations, Var - Dhauli.

Sl. No.	Treatment Code	Plant height (cm)	No. of pods/plant	Pod length (cm)	Seeds per pod	100-seed weight (g)	Single plant yield (g)
1	E ₁	49.75	22.30	7.90**	5.40**	3.76**	4.85**
2	E ₂	47.85	29.20**	6.70	5.30**	4.20**	6.60**
3	S ₁	40.60*	16.70	6.60	5.35**	3.81**	3.55
4	S ₂	38.85*	28.20**	7.60**	4.90**	3.98**	5.55**
5	N ₁	37.75*	15.90*	6.20	4.86**	3.14	2.50*
6	N ₂	39.20*	17.70	6.90	3.75*	2.87*	2.00*
7	E ₂ S ₂	50.40**	17.80	6.40	4.25	2.70*	2.00*
8	E ₂ N ₂	51.10**	14.40*	6.30	3.70*	4.32**	2.30*
9	S ₂ N ₂	48.40	17.50	6.60	4.90**	2.90*	2.60*
10	E ₁ S ₁	45.05	29.70**	9.10**	5.50**	4.58**	8.05**
11	E ₁ N ₁	38.25*	18.00	6.50	3.50*	2.90*	2.00*
12	S ₁ N ₁	46.65	15.90*	6.20	4.25	3.43	3.00
13	E ₂ S ₁	48.30	17.60	9.30**	3.80*	3.03**	2.00*
14	E ₂ N ₁	50.70**	23.60**	6.90	5.70**	4.25**	6.10**
15	S ₂ N ₁	44.10*	26.70**	6.80	4.90**	4.05**	6.60**
16	E ₁ S ₂	40.40*	16.70	6.20	3.60*	3.18	2.00*
17	E ₁ N ₂	40.35*	17.20	6.80	4.50	2.89*	2.60*
18	S ₁ N ₂	40.95*	29.46**	7.60**	5.90**	4.21**	7.30**
19	Control	46.10	18.80	6.40	4.60	3.40	3.50
	C.D.(0.05)	4.19	3.83	1.07	0.21	0.32	0.87

* Significant decrease at P = 0.05

** Significant increase at P = 0.05

Table 15. Mean values of quantitative traits in M₂ generation of different mutagen treated populations, Var - Nayagarh local.

Sl. No.	Treatment Code	Plant height (cm)	No. of pods/plant	Pod length (cm)	Seeds per pod	100-seed weight (g)	Single plant yield (g)
1	E ₁	26.30*	20.00**	6.30	5.00**	3.89	4.20**
2	E ₂	36.65*	21.30**	6.30	5.10**	3.47*	3.80**
3	S ₁	37.35*	18.70	7.20	4.40	4.29**	3.50
4	S ₂	35.50*	15.90	5.60	3.30*	2.92*	4.10**
5	N ₁	41.50	21.60**	6.60	4.30	3.88	3.75
6	N ₂	42.60	24.50**	7.90**	4.30	3.92	4.00**
7	E ₂ S ₂	36.20*	21.60**	6.10	4.00	2.99*	2.70*
8	E ₂ N ₂	40.50	11.30*	6.10	3.80	4.12**	2.85*
9	S ₂ N ₂	40.35	14.20	5.80	4.20	2.79*	1.75*
10	E ₁ S ₁	31.50*	22.30**	5.20*	3.10*	2.89*	4.30**
11	E ₁ N ₁	31.65*	13.70	5.40*	3.30*	2.75*	1.35*
12	S ₁ N ₁	36.90*	13.50	6.80	3.60*	3.44*	1.85*
13	E ₂ S ₁	30.65*	16.40	6.10	3.50*	2.33*	1.40*
14	E ₂ N ₁	39.65	17.20	6.70	4.60*	3.55*	3.50
15	S ₂ N ₁	38.90	16.70	6.50	5.30**	3.64	4.15**
16	E ₁ S ₂	40.30	15.20	6.50	3.30*	3.04*	1.70*
17	E ₁ N ₂	35.95*	13.90	6.00	4.40	3.17*	2.10*
18	S ₁ N ₂	38.30*	24.60**	7.10	5.60**	4.42**	6.50**
19	Control	42.60	16.50	6.50	4.20	3.84	3.30
	C.D.(0.05)	4.14	3.60	0.96	0.55	0.21	0.40

* Significant decrease at P = 0.05

** Significant increase at P = 0.05

4.3.6 Yield/plant

The treated populations of Dhauri varied from 2.00 to 8.05 with respect to yield/plant (g) as against 3.50 in the control. In Nayagarh local, yield/plant of the treated populations varied from 1.35 to 6.50 as against 3.30 in the control. A significant increase in yield/plant over control was observed in E_1 , E_2 , S_2 , E_1S_1 , E_2N_1 , S_2N_1 and S_1N_2 treated populations of Dhauri. Populations treated with E_1 , E_2 , S_2 , N_2 , E_1S_1 , S_2N_1 and S_1N_2 exhibited a significant increase in this character over control in Nayagarh local. All the treated populations of both varieties showed change of mean both in positive and negative directions; highest positive shift being in S_1N_2 treatment and negative shift in E_2S_1 treatment in both the varieties.

4.4 POPULATION VARIANCE

4.4.1 Plant height

In the treated populations of both the varieties, the variance was enlarged over control. The variance ranged from 13.59 to 40.48 in Dhauri and 12.32 to 24.62 in Nayagarh local as against 13.25 and 12.04 in the respective controls (Tables 16 and 17). Increase in variance was significant in all the treated populations of Dhauri except E_2 . Except S_1N_1 and S_2N_1 , all other treated populations of Nayagarh local showed increase in variance. When single mutagenic treatment was considered, high variances was observed in NG treated populations followed by SA and EMS in Dhauri, while, it was highest in EMS treated populations followed by SA and NG in Nayagarh local.

4.4.2 Pods per plant

In the treated populations, the variance for pods per plant ranged from 11.02 to 24.51 and 10.65 to 24.45 in Dhauli and Nayagarh local respectively. The variance for control populations was 9.67 for Dhauli and 8.11 for Nayagarh local. All the treated populations of Dhauli except S_2 and N_2 showed significant increase in variance. In Nayagarh local, the variance of all the treated populations was significantly higher than control. In both the varieties, increase in variance was more in combine treated populations as compared to the single mutagenic treatment. Single mutagenic treatment was observed to be dose dependant. On an average the variance was highest in E_1N_1 and E_2S_2 treated populations of Dhauli and Nayagarh local respectively.

4.4.3 Seeds per pod

The variance of seeds per pod of the treated populations varied from 10.52 to 20.71 in Dhauli and 10.29 to 22.15 in Nayagarh local as against 5.85 and 4.93 in the respective controls. There was a significant increase in variance of the treated populations as compared to control in both the varieties. In general, the variance observed in this character in combined treatments was higher than the single mutagenic treatment. When single mutagenic treatment was considered, populations treated with NG recorded highest variance followed by SA and EMS in both the varieties. The increase in variance was observed to be dose dependent, except EMS treated populations of Nayagarh local.

Table 16. Treatment population coefficient of variation for different quantitative traits in M₂ generation. Var - Dhuli.

Sl. No.	Treatment Code	Plant height	Pods/plant	Seeds/pod	100-seed weight (g)	Single plant yield (g)
1	E ₁	16.86*	13.51*	12.86*	25.86*	12.38*
2	E ₂	13.59	13.17*	10.62*	22.98*	11.64*
3	S ₁	18.03*	18.62*	15.26*	10.00*	17.75*
4	S ₂	23.18*	11.02	10.57*	25.97*	10.34*
5	N ₁	25.98*	19.51*	18.88*	14.15*	11.64*
6	N ₂	40.48*	11.15	11.57*	19.43*	11.25*
7	E ₂ S ₂	20.98*	17.5*	15.33*	17.94*	17.13*
8	E ₂ N ₂	25.82*	19.61*	15.40*	21.90*	26.90*
9	S ₂ N ₂	27.47*	19.03*	19.00*	13.10*	19.89*
10	E ₁ S ₁	32.34*	17.53*	17.72*	24.50*	17.87*
11	E ₁ N ₁	19.15*	22.97*	19.64*	15.44*	19.82*
12	S ₁ N ₁	23.03*	19.61*	12.17*	11.27*	11.39*
13	E ₂ S ₁	16.77*	18.02*	16.19*	12.01*	17.20*
14	E ₂ N ₁	17.04*	17.26*	12.61*	19.27*	17.35*
15	S ₂ N ₁	15.03*	24.51*	20.71*	11.65*	21.15*
16	E ₁ S ₂	18.74*	17.42*	11.23*	13.60*	17.97*
17	E ₁ N ₂	14.77*	20.14*	20.27*	15.83*	19.86*
18	S ₁ N ₂	14.94*	21.72*	20.38*	24.67*	23.77*
19	Control	13.26	9.67	5.85	4.50	6.15
	C.D.(0.05)	1.27	3.43	1.34	1.70	0.72

* Significant increase in variance than control at P = 0.05

Table 17. Treatment population coefficient of variation for different quantitative traits in M₂ generation. Var - Nayagarh local.

Sl. No.	Treatment Code	Plant height	Pods/plant	Seeds/pod	100-seed weight (g)	Single plant yield (g)
1	E ₁	17.56*	11.38*	10.29*	23.35*	12.81*
2	E ₂	20.10*	10.65*	10.38*	22.11*	11.85*
3	S ₁	19.85*	15.31*	10.51*	16.17*	14.86*
4	S ₂	17.05*	11.96*	10.48*	19.57*	10.02*
5	N ₁	17.14*	18.33*	14.03*	25.12*	18.77*
6	N ₂	15.09*	17.36*	11.41*	24.08*	18.34*
7	E ₂ S ₂	23.40*	24.45*	17.94*	25.83*	19.22*
8	E ₂ N ₂	22.22*	19.14*	15.35*	7.06*	17.54*
9	S ₂ N ₂	15.60*	19.27*	15.41*	9.69*	16.54*
10	E ₁ S ₁	17.43*	15.25*	18.71*	22.42*	17.88*
11	E ₁ N ₁	16.66*	11.33*	12.40*	11.20*	12.73*
12	S ₁ N ₁	12.32	18.63*	15.25*	9.89*	16.91*
13	E ₂ S ₁	23.24*	19.01*	13.89*	28.89*	16.72*
14	E ₂ N ₁	19.82*	18.36*	19.07*	8.35*	19.68*
15	S ₂ N ₁	13.50	16.58*	19.20*	21.05*	20.90*
16	E ₁ S ₂	20.98*	16.98*	15.88*	7.67*	19.38*
17	E ₁ N ₂	17.73*	20.52*	18.33*	7.66*	21.78*
18	S ₁ N ₂	24.62*	18.11*	22.15*	18.01*	21.84*
19	Control	12.04	8.11	4.93*	4.49	5.96
	C.D.(0.05)	1.47	0.82	1.86	1.30	1.40

* Significant increase in c v than control at P = 0.05

4.4.4 100-seed weight

The treated populations of both the varieties showed significant increase in variance of 100-seed weight over the control. Variance of 100-seed weight in the treated populations ranged from 10.00 to 25.97 in Dhauli and 7.06 to 28.89 in Nayagarh local as against 4.50 and 4.49 of the respective controls. In single mutagenic treatments, EMS treated populations of Dhauli recorded highest variance followed by SA and NG while, the variance was more in NG treated populations followed by EMS and SA in Nayagarh local. The variance of this character appeared to be low on many of the combine treated populations as compared to single mutagenic treated populations.

4.4.5 Yield per plant

Variance of single plant yield (g) ranged from 10.34 to 26.90 in Dhauli and 10.02 to 21.78 in Nayagarh local as against 6.15 and 5.96 in the controls respectively. The treated populations of both the varieties showed significant increase in variance over control. Increase in variance was observed in the most of the combined treatment populations than single treated populations of both the varieties. In Dhauli populations treated with SA, EMS and NG recorded higher variance in descending order. In Nayagarh local, populations treated with NG showed highest variance followed by SA and EMS.

4.5 CORRELATION OF M_1 PARAMETERS WITH MACRO-MUTATION FREQUENCY IN M_2

Correlation of M_1 parameters with M_2 mutation frequency was estimated to assess the bearing of M_1 parameters on induction of macro-mutations in M_2 generation and presented in

631.53

2658

NG 100

Table 18. Correlation Coefficient (r) of Chlorophyll mutation frequency in M₂ with different M₁ parameters.

M ₁ Parameters	Chlorophyll mutation frequency (%) in M ₂	
	Dhauri	Nayagarh local
Germination %	0.4013	0.5396*
Survival %	0.6642**	0.5101*
Seedling height (cm)	0.4504	0.7435**
Pollen sterility (%)	0.5822**	0.6825**
Seeds sterility (%)	0.7161**	0.5658**

* Significant at 0.05

** Significant at 0.01

Table 19. Correlation Coefficient (r) of M₁ parameters with M₂ mutations frequency in some quantitative traits.

M ₁ Parameters	Population variance of M ₂ Generation					
	Dhauri			Nayagarh local		
	Pods/plant	Seeds/pod	Single plant yield (g)	Pods/plant	Seeds/pod	Single plant yield (g)
Germination %	0.4948*	0.5386*	0.6521**	0.5630**	0.3653	0.656**
Survival %	0.6562**	0.752**	0.8069**	0.5962**	0.6439**	0.5865**
Seedling height (cm)	0.4398	-0.3785	0.3203**	-0.6279**	0.5832**	0.5178*
Pollen sterility (%)	0.6506**	0.7242**	0.4533*	0.7185**	0.4991*	0.3594
Seeds sterility (%)	0.8947**	0.9377**	0.3899	0.8621**	0.6406**	0.7760**

* Significant at 0.05

** Significant at 0.01

Table 18. The M_1 parameters like survival, pollen sterility and seed sterility in both the varieties showed positive significant correlation with M_2 chlorophyll mutation frequency. Of these, seed sterility showed highly significant association in Dhauli, whereas it was high for pollen sterility in Nayagarh local.

The inter relationship between the effect of mutagenic treatment on M_1 generation and induced variability observed for yield and yield components like seeds/pod, pods/plant and yield/plant in M_2 generation was worked out and presented in Table 19. A negative association between seedling height and seeds/pod in Dhauli and pods/plant in Nayagarh local was observed. The correlation between other characters of both the varieties was positive. In both the varieties, seed sterility and pollen sterility showed highly significant correlation with M_2 variances of the above mentioned three characters.

4.6 EFFICIENCY OF SINGLE AND COMBINED MUTAGENIC TREATMENT

For extensive use of mutants in breeding, the high efficiency of their action is essential. In mutation breeding, the usefulness of any mutagen used either single or in combination depends not only on its mutagenic efficiency but also the degree of biological damage it could possibly cause.

The result presented in the Tables 20 and 21 indicated the efficiency of mutagens based on different biological damage. It was noted that NG was more efficient than other two chemicals on the basis of survival. The order of mutagens based on the efficiency judged by the frequency of mutations in relation to undesirable biological effect, was NG > SA > EMS in

Table 20. Relative efficiency of single and combine treatment of mutagen in greengram, Var - Dhauli.

Sl. No.	Treatment Code	Chlorophyll mutagen (%)	Survival (%)	Efficiency	Pollen Sterility (%)	Efficiency	Seed Sterility (%)	Efficiency
1	E ₁	1.47	68.0	0.021	10.2	0.144	9.3	0.158
2	E ₂	1.65	70.0	0.023	9.3	0.177	8.5	0.194
3	S ₁	1.39	60.0	0.023	12.8	0.108	10.3	0.135
4	S ₂	1.77	66.0	0.027	10.3	0.171	7.5	0.236
5	N ₁	1.71	58.0	0.029	17.2	0.099	11.9	0.143
6	N ₂	2.00	66.0	0.030	12.3	0.163	9.2	0.217
7	E ₂ S ₂	2.84	56.0	0.051	16.0	0.177	10.3	0.275
8	E ₂ N ₂	2.74	50.0	0.054	13.5	0.203	11.5	0.238
9	S ₂ N ₂	3.11	50.0	0.062	13.0	0.240	11.7	0.265
10	E ₁ S ₁	2.61	60.0	0.043	12.9	0.202	10.7	0.243
11	E ₁ N ₁	2.58	64.0	0.043	12.1	0.213	12.1	0.213
12	S ₁ N ₁	2.90	66.0	0.044	16.1	0.180	12.1	0.240
13	E ₂ S ₁	2.21	60.0	0.036	15.3	0.153	10.4	0.225
14	E ₂ N ₁	2.34	54.0	0.043	17.9	0.147	10.9	0.241
15	S ₂ N ₁	2.30	50.0	0.046	17.9	0.128	12.0	0.191
16	E ₁ S ₂	2.59	56.0	0.046	14.2	0.182	10.5	0.246
17	E ₁ N ₂	2.68	52.0	0.051	14.2	0.189	12.5	0.214
18	S ₁ N ₂	2.91	44.0	0.066	17.3	0.168	12.0	0.242
19	Control	Nil	-	-	0.3	-	0.7	-

Table 21. Relative efficiency of single and combine treatment of mutagens in greengram, Var - Nayagarh local.

Sl. No.	Treatment Code	Chlorophyll mutagen (%)	Survival (%)	Efficiency	Pollen Sterility (%)	Efficiency	Seed Sterility (%)	Efficiency
1	E ₁	1.29	60.0	0.021	12.3	0.104	9.7	0.132
2	E ₂	1.51	70.3	0.022	10.1	0.149	8.1	0.186
3	S ₁	1.04	56.0	0.018	12.3	0.084	10.8	0.096
4	S ₂	1.17	70.0	0.017	10.7	0.109	9.1	0.128
5	N ₁	1.36	57.0	0.020	13.0	0.104	13.2	0.103
6	N ₂	1.81	61.7	0.030	12.0	0.150	11.5	0.157
7	E ₂ S ₂	3.01	56.3	0.053	18.1	0.166	13.7	0.219
8	E ₂ N ₂	2.74	50.5	0.054	17.3	0.158	12.7	0.215
9	S ₂ N ₂	3.44	53.4	0.064	15.3	0.224	12.9	0.266
10	E ₁ S ₁	3.41	53.3	0.063	12.4	0.275	11.8	0.289
11	E ₁ N ₁	3.19	62.5	0.051	13.2	0.241	9.5	0.335
12	S ₁ N ₁	2.90	57.3	0.051	14.2	0.204	11.3	0.256
13	E ₂ S ₁	2.44	60.0	0.040	12.5	0.195	11.7	0.208
14	E ₂ N ₁	2.76	58.3	0.047	14.0	0.197	14.2	0.194
15	S ₂ N ₁	3.28	51.2	0.064	12.7	0.258	10.8	0.303
16	E ₁ S ₂	2.39	51.8	0.046	12.4	0.192	10.7	0.223
17	E ₁ N ₂	2.41	52.3	0.046	12.6	0.191	12.1	0.199
18	S ₁ N ₂	3.15	56.3	0.055	12.7	0.248	12.4	0.254
19	Control	-	-	-	0.43	-	1.0	-

survival; EMS > SA > NG in pollen sterility and SA > NG > EMS for seed sterility in Dhauri. In Nayagarh local, it was NG > EMS > SA for survival; EMS > NG > SA for pollen sterility and EMS > SA > NG for seed sterility. The result indicated that SA & EMS was more efficient mutagenes for Dhauri and Nayagarh local respectively as far as single mutagenic treatment was concerned. The combine treatments of mutagens were found to be more efficient than single treatment in both the varieties.

4.7 M₃ GENERATION

From the M₂ populations of each treatments of both the varieties twelve plants were selected on the basis of higher yield and were carried forward to grow M₃ generations. Seeds of these plants were grown in compact family block design with three replications. The treatments were taken in the main plot and progenies in the sub-plots. Two separate trials for both the varieties were conducted. The average yield/plant of the progenies were estimated and details of which are in Tables 22 and 23.

The average yield/plant (g) obtained in control plants of Dhauri was 2.83. The treated populations of E₁, E₂, S₂, N₁, N₂, S₂N₂, E₁S₁, E₂N₁, S₂N₁, E₁S₂ and S₁N₂ showed high significant increase in mean yield over control. The average yield/plant in Nayagarh local was 2.27. The treatments those gave significant higher yield/plant over control were E₁, E₂, S₁, S₂, N₁, N₂, E₂S₂, E₂N₂, S₂N₂, E₁S₁, E₂N₁, S₂N₁, E₁N₂ and S₁N₂.

There exists significant difference among treatments of both the varieties with respect to yield/plant. In both the varieties, the progenies of E₁S₁ yielded higher yield/plant

than other treatments. In Dhauli, the mean yield/plant of the treatments varied from 2.83 in control to 4.96 in treatment S_1N_2 . The observed variation in Nayagarh local with respect to yield/plant varied from 2.28 in control to 4.98 in E_1S_1 .

Analysis of variance for between progeny within treatment indicated that within treatment, progeny variation was significant for 14 in Dhauli and 13 in Nayagarh local. The range of variation within treatment with respect to yield/plant was wider as compared to respective controls. The progeny difference was significant for E_2 , S_1 , S_2 , N_1 , N_2 , E_2S_2 , E_2N_2 , S_2N_2 , E_1S_1 , E_1N_1 , E_2N_1 , S_2N_1 , E_1S_2 and S_1N_2 in Dhauli and for E_1 , E_2 , S_2 , N_2 , E_2S_2 , E_2N_2 , E_1S_1 , E_1N_1 , S_1N_1 , E_2N_1 , E_1N_2 , E_1S_2 , and S_1N_2 in Nayagarh local.

The progenies having 50 % higher yield/plant over the superior progeny of the control was considered as the base for further selection. In Dhauli, such superior progenies were obtained in treatments E_1 (5), E_2 (8), N_2 (4) E_1S_1 (11), E_2N_1 (6) and S_1N_2 (12). Of these 46 superior progenies obtained in Dhauli, only 17 (36.95 %) were from single mutagenic treatments and rest 29 (63.05 %) were from combined treatments. Further, it was observed that EMS mutagen administered either singly or in combination produced high percentage of superior progenies than the other two mutagens. From the seventeen superior progenies selected amongst single mutagenic treatments, 13 (76.5 %) were selected from EMS treatments only and the rest were from NG treatment. Higher percentage of superior progenies obtained from combine treated population indicated the better performance of combine treatment of mutagens.

Table 22. Mean Single Plant yield of different M₃ Progenies of different mutagenic treatments, Var - Dhaulii.

Progeny No.	Mutagenic treatments																Control		
	E ₁	E ₂	S ₁	S ₂	N ₁	N ₂	E ₂ S ₂	E ₂ N ₂	S ₂ N ₂	E ₁ S ₁	E ₁ N ₁	S ₁ N ₁	E ₂ S ₁	E ₂ N ₁	S ₂ N ₁	E ₁ S ₂		E ₁ N ₂	S ₁ N ₂
1	4.38	4.33	2.08	3.18	3.93	4.32	2.42	2.66	2.91	4.23	2.41	2.76	2.35	4.27	4.04	3.13	2.83	5.01	2.84
2	4.17	4.15	2.13	3.28	9.39	4.06	2.59	2.76	2.89	4.51	2.60	2.91	2.47	4.30	3.93	3.12	2.68	5.03	2.86
3	4.25	4.36	2.09	3.42	3.65	4.14	2.61	2.58	3.00	4.68	2.85	2.88	2.46	4.08	3.98	3.23	2.73	4.85	2.83
4	4.31	4.36	2.14	3.28	3.51	4.16	2.47	2.39	2.10	4.93	2.49	2.80	2.65	4.40	4.17	3.10	2.74	4.93	2.81
5	4.19	4.07	1.95	3.39	3.90	4.34	2.68	2.48	2.98	4.90	2.51	2.86	2.64	4.35	4.18	3.03	2.87	4.93	2.82
6	4.24	4.37	1.97	3.49	3.65	4.16	2.49	2.22	2.67	5.40	2.45	2.89	2.58	4.28	3.92	3.11	2.83	4.94	2.85
7	4.23	4.27	2.12	3.35	3.86	4.47	2.43	2.62	2.97	5.01	2.71	2.96	2.40	4.28	3.85	3.08	2.68	5.11	2.83
8	4.19	4.33	2.05	3.52	3.76	4.32	2.53	2.69	3.02	5.20	2.47	2.86	2.55	4.23	4.20	3.08	2.57	4.93	2.83
9	4.21	4.43	2.05	3.11	3.92	3.99	2.52	2.65	2.70	4.94	2.47	2.78	2.46	4.12	3.98	3.11	3.13	4.97	2.84
10	4.39	4.27	2.31	3.15	3.88	3.97	2.45	2.21	2.86	4.85	2.50	2.71	2.43	4.34	4.21	3.12	2.80	5.09	2.83
11	4.23	4.30	1.95	3.26	3.86	3.99	2.44	2.54	2.98	4.93	2.47	2.83	2.86	4.39	4.17	3.18	2.80	4.86	2.81
12	4.35	4.37	1.97	3.28	3.92	4.12	2.49	2.54	2.28	4.97	2.62	2.98	2.55	4.38	3.95	3.14	2.79	4.96	2.82
Mean	4.23	4.28	2.06	3.31	3.77	4.11	2.51	2.61	2.88	4.85	2.54	2.82	2.51	4.28	4.10	3.12	2.79	4.96	2.83
C.D. (0.05)	N.S.	0.012	0.071	0.13	0.18	0.05	0.13	0.19	0.11	0.15	0.12	N.S.	N.S.	0.15	0.12	0.08	N.S.	0.12	N.S.

Table 23. Mean Single Plant yield of different M₃ Progenies of different mutagenic treatments Var - Nayagarh local

Progeny No.	Mutagenic treatments																Control		
	E ₁	E ₂	S ₁	S ₂	N ₁	N ₂	E ₂ S ₂	E ₂ N ₂	S ₂ N ₂	E ₁ S ₁	E ₁ N ₁	S ₁ N ₁	E ₂ S ₁	E ₂ N ₁	S ₂ N ₁	E ₁ S ₂		E ₁ N ₂	S ₁ N ₂
1	3.92	3.30	3.66	3.93	3.13	3.82	2.95	2.95	3.82	4.83	1.43	1.96	1.63	3.85	2.37	2.02	2.37	4.25	2.28
2	4.02	3.43	3.51	4.12	3.15	4.05	2.93	3.06	3.83	5.11	1.46	1.95	1.67	4.14	2.39	2.01	2.38	4.00	2.28
3	4.09	3.55	3.31	3.85	3.13	4.08	2.87	3.13	4.13	5.11	1.43	2.03	1.69	4.14	2.72	2.02	2.31	4.16	2.26
4	4.01	3.55	3.79	4.10	3.25	3.98	2.93	3.19	3.82	4.88	1.40	2.07	1.57	3.96	2.32	1.99	2.38	4.08	2.26
5	4.19	3.19	3.46	3.92	3.11	3.85	2.77	3.04	3.78	5.04	1.37	1.97	1.61	3.96	2.30	2.03	2.29	4.02	2.27
6	4.25	3.10	4.12	3.86	3.04	3.88	2.85	2.88	3.88	4.95	1.36	1.95	2.63	3.96	2.43	1.99	2.29	4.02	2.70
7	3.97	3.40	4.62	3.83	3.09	4.14	2.93	2.99	4.15	5.13	1.40	2.10	1.62	3.95	2.31	1.97	2.30	4.11	2.27
8	4.08	3.19	3.60	3.98	3.06	4.18	2.94	3.07	3.92	5.02	1.46	1.88	1.63	3.97	2.40	1.99	2.28	4.11	2.27
9	3.94	3.17	3.63	4.14	2.99	3.93	2.84	3.15	4.06	5.10	1.40	1.95	1.59	3.58	2.21	2.02	2.39	4.10	2.26
10	4.10	3.21	3.35	4.10	3.11	4.23	2.92	2.84	3.85	4.75	1.40	2.01	1.65	3.94	2.51	2.04	2.39	4.20	2.28
11	4.12	3.25	4.07	3.96	3.24	4.32	2.92	2.80	3.85	4.75	1.42	2.01	1.62	3.84	2.54	2.04	2.44	4.24	2.27
12	4.01	3.39	3.50	4.14	3.13	4.33	2.71	2.89	3.58	4.93	1.43	2.04	1.65	4.15	2.68	2.02	2.53	4.24	2.28
Mean	4.06	3.31	3.52	3.97	3.13	4.05	2.88	2.99	3.82	4.98	1.42	1.99	1.71	3.96	2.43	2.01	2.36	4.13	2.27
C.D. (0.05)	0.16	0.08	N.S.	0.16	N.S.	0.11	0.09	0.12	N.S.	0.06	0.05	0.06	N.S.	0.27	N.S.	0.04	0.05	0.08	N.S.

The yield/plant (g) of the superior progeny of the control in Nayagarh local was 2.70. The treatment progenies yielding 50 % higher yield than the superior progenies of control were 46 in number. They were from the treatments E_1 (6), S_1 (3), S_2 (5), N_2 (7), S_2N_2 (3), E_1S_1 (12) and S_1N_2 (10). Of the 46 superior progenies obtained, 25 (54.35 %) were from combined mutagenic treatments and 21 (45.65 %) were from single mutagenic treatments, indicating the better performance of combine treatment of mutagens. The highest percentage of superior progenies were obtained in SA treatment followed by NG and EMS respectively in descending order irrespective of their mode of treatments.

4.8 M_4 GENERATION

Forty six selected progenies, on the basis of higher yield, were selected from each variety and were carried forward to M_4 generation. They were numbered serially with prefix of either E, S or N for EMS, SA and NG respectively in addition to D for Dhauri and N for Nayagarh local. Two separate trials were conducted for the two varieties. Forty six selected mutants for both the varieties were evaluated against their respective controls in RBD with three replications. The results pertaining to this study are presented in the following sections.

4.8.1 Mean of the quantitative characters

The mean performances of the eight quantitative traits of the selected mutant cultures of both the varieties are presented in Tables 24 and 25.

Days to maturity : The variation created in maturity (days) due to induction of mutation in the mutant cultures was ranged

from 48 to 65 in Dhauli and 51.98 to 66.33 in Nayagarh local as against 60 and 67 in the respective controls. In Dhauli three mutant cultures matured significantly late from the control whereas thirty mutants matured significantly early. In Nayagarh local forty mutant cultures were significantly early in maturity as compared to control.

Plant height : The plant height (cm) of the mutant cultures varied from 20.37 to 61.23 in Dhauli and 30.17 to 41.00 in Nayagarh local, as against 47.4 and 48.2 in the respective controls. There was a significant reduction in plant height in all the mutant cultures of Nayagarh local from the control. In Dhauli, thirty one cultures showed significant reduction in plant height while five cultures exhibited significant increase in plant height.

Number of branches/plant : The number of branches/plant of the mutant cultures ranged from 1.13 to 1.83 in Dhauli and 1.37 to 1.90 in Nayagarh local. The control plants showed 1.17 and 1.57 branches/plant respectively in Dhauli and Nayagarh local. Thirty mutant cultures of Dhauli showed significant increase in branches/plant from the control. In Nayagarh local, sixteen cultures showed significant increase and five cultures observed significant decrease in branches/plant, when compared with the control.

Pods/plant : The range of variability in pods/plants of the mutant cultures of Dhauli and Nayagarh local was from 21.67 to 48.94 and 18.48 to 38.82 respectively as against 21.67 and 18.5 of the respective controls. Forty five mutant cultures from both the varieties showed significant increase in pods/plant

Table 24. Mean of Eight characters of mutant cultures , Var Dhauli, M₄ generation.

Sl. No.	Name of Cultures	50%Flowering (days)	Maturity (days)	Plant height (cm)	Branches/ Plant	Pods/ Plant	Seeds / Pod	100-Seed weight (g)	Plant yield (g)
1	DE ₁ -1	47.4**	60.3	29.4*	1.30**	25.87**	4.61**	3.64	4.07**
2	DE ₁ -3	42.0	59.3	28.5*	1.43**	48.94**	3.05	3.09	4.63**
3	DE ₁ -4	39.7*	56.3*	31.3*	1.73**	24.61**	4.58**	3.89	4.56**
4	DE ₁ -10	44.3**	61.0	30.8*	1.30**	28.00**	4.78**	3.71	4.58**
5	DE ₁ -12	41.7	48.0*	36.6*	1.23	40.10**	4.07	2.61	4.79**
6	DE ₂ -1	46.7**	64.7**	41.0*	1.13	25.60**	4.61	3.71	4.13**
7	DE ₂ -3	45.0**	65.0**	37.6*	1.27	22.40	4.86**	3.80	4.36**
8	DE ₂ -4	42.2	61.7*	42.3*	1.30**	26.60**	4.56	3.94**	4.27**
9	DE ₂ -6	43.0	61.2*	45.3*	1.23	28.50**	3.96	3.90	4.28**
10	DE ₂ -8	40.4	61.7*	48.0	1.33**	24.90**	3.95	3.68	4.07**
11	DE ₂ -9	41.3	59.7	47.8	1.40**	31.30**	3.99	3.71	4.14**
12	DE ₂ .11	40.3	58.3	47.6	1.40**	30.70**	4.01	2.62	3.86**
13	DE ₂ -12	41.3	58.0	47.0	1.20	30.80**	3.67	3.59	4.17**
14	DN ₂ -1	47.7**	57.7*	45.5	1.33**	30.90**	4.03	3.44	4.27**
15	DN ₂ -5	39.9*	54.1*	38.3*	1.57**	23.80**	4.85**	3.66	4.18**
16	DN ₂ -7	40.7	57.3*	58.0**	1.43**	41.10**	3.74	3.05	4.21**
17	DN ₂ -8	40.4	58.0	56.9**	1.33**	45.20**	3.02	3.60	4.61**
18	DE ₁ S ₁ -1	40.7	59.0	50.3**	1.23	43.20**	3.02	3.61	4.61**
19	DE ₁ S ₁ -2	40.0*	57.3*	48.8*	1.27	41.60**	4.05	3.02	4.95**
20	DE ₁ S ₁ -3	35.3*	51.2*	37.5*	1.23	22.20	4.75**	2.84	3.85**
21	DE ₁ S ₁ -4	35.3*	51.3*	38.8*	1.63**	27.60**	4.84**	3.64	4.47**
22	DE ₁ S ₁ -5	35.3*	57.3*	61.2**	1.60**	27.40**	4.53	4.32**	5.29**
23	DE ₁ S ₁ -6	36.7*	55.0*	48.9*	1.70**	39.20**	4.06	3.41	5.26**
24	DE ₁ S ₁ -7	36.7*	54.4*	53.8**	1.67**	25.60**	4.93**	4.15**	5.25**
25	DE ₁ S ₁ -9	36.3*	50.7*	41.5*	1.57**	28.70**	0.33	3.87	4.79**
26	DE ₁ S ₁ -10	36.0*	50.3*	40.8*	1.67**	26.00**	5.01**	3.65	4.66**
27	DE ₁ S ₁ -11	34.3*	52.3*	30.5*	1.57**	26.80**	4.96**	3.72	5.08**
28	DE ₁ S ₁ -12	32.4*	49.3*	48.2	1.43**	28.70**	4.72**	3.73	5.09**
29	DE ₂ N ₁ -2	39.1*	56.0*	48.2	1.37**	28.30**	4.72**	5.08**	4.20**
30	DE ₂ N ₁ -4	40.1*	57.0*	39.3*	1.17	34.70**	3.47	3.41	4.09**
31	DE ₂ N ₁ -5	38.8*	56.7*	39.3*	1.17	24.70**	4.76**	3.68	4.26**
32	DE ₂ N ₁ -8	40.3*	59.0*	33.5*	1.13	30.50**	4.65**	3.62	4.65**
33	DE ₂ N ₁ -12	40.6	62.7**	37.3*	1.20	28.80**	4.71**	3.61	4.29**
34	DE ₂ N ₁ -12	41.7	60.3	37.4*	1.23	25.30**	4.78**	3.65	4.65**
35	DS ₁ N ₂ -1	34.6*	51.2*	32.1*	1.67**	27.50**	4.82**	4.04**	5.09**
36	DS ₁ N ₂ -2	34.6*	51.1*	35.2*	1.60**	25.50**	4.65**	4.27**	5.03**
37	DS ₁ N ₂ -3	33*	50.5*	29.1*	1.27	25.80**	4.84**	3.95**	4.66**
38	DS ₁ N ₂ -4	34.5*	50.1*	35.2*	1.23	27.40**	4.63**	3.81	4.74**
39	DS ₁ N ₂ -5	35.8*	52.2*	34.6*	1.17	28.50**	4.71**	3.63	4.83**
40	DS ₁ N ₂ -6	35.3*	51.0*	37.7*	1.63**	30.40**	4.26	3.77	4.69**
41	DS ₁ N ₂ -7	34.8*	51.2*	38.5*	1.77**	34.50**	5.11**	3.56	5.20**
42	DS ₁ N ₂ -8	36.1*	52.8*	39.1*	1.77**	34.20**	4.03	3.27	5.11**
43	DS ₁ N ₂ -9	34.4*	52.0*	40.2*	1.83**	37.70**	4.24	3.23	5.06**
44	DS ₁ N ₂ -10	33.3*	50.1*	39.1*	1.77**	35.00**	4.00	4.04**	5.00**
45	DS ₁ N ₂ -11	34.3*	54.9*	41.3*	1.77**	24.70**	4.80**	4.17**	4.97**
46	DS ₁ N ₂ -12	35.3*	50.8*	35.7*	1.73**	37.50**	4.02	4.20**	5.21**
47	DC	42.7	60.0	47.4	1.17	21.70	4.51	3.27	3.55
CD(0.05)		1.38	2.08	1.86	0.12	0.93	0.06	0.67	0.11

* Significant decrease at 0.05

** Significant increase at 0.05

Table 25. Mean of Eight characters of mutant cultures , Var Nayagarh local, M₄ generation.

Sl. No.	Name of Cultures	50% Flowering (days)	Maturity (days)	Plant height (cm)	Branches/ Plant	Pods/ Plant	Seeds / Pod	100-Seed weight (g)	Plant yield (g)
1	NE ₁ -3	47.67**	62.7*	31.3*	1.63	26.20**	4.57**	3.32**	3.99**
2	NE ₁ -5	50.90**	65.4	32.3*	1.67	26.50**	4.88*	3.31**	4.10**
3	NE ₁ -6	42.70*	57.9*	37.7*	1.33*	28.60**	4.22	3.37**	4.05**
4	NE ₁ -8	44.30*	61.5*	41.1*	1.40*	30.60**	4.21	3.38**	3.39**
5	NE ₁ -10	45.00	58.7*	38.7*	1.40*	26.60**	4.39**	3.40**	4.04**
6	NE ₁ -11	39.00*	56.3*	41.0*	1.77**	28.30**	4.01	3.41**	3.90**
7	NS ₁ -6	49.00	66.2	33.1*	1.83**	25.70**	4.70**	3.71**	4.50**
8	NS ₁ -7	43.00*	62.3*	30.7*	1.83**	20.40**	4.52**	3.62**	3.47**
9	NS ₁ -11	50.70**	65.3*	32.2*	1.93**	18.80*	4.68**	3.71**	3.29**
10	NS ₂ -2	48.30**	60.1*	33.3*	1.73**	24.20**	4.88**	3.98**	4.11**
11	NS ₂ -4	41.30*	59.3*	38.7*	1.63	28.30**	5.80**	3.01	4.09**
12	NS ₂ -9	44.30*	64.7*	33.7*	1.60	25.60**	4.31*	3.63**	4.01**
13	NS ₂ -10	44.40	63.9*	41.0*	1.53	30.20**	4.32*	3.03	4.05**
14	NS ₂ -12	45.30	63.4*	33.2*	1.67	20.60**	4.85**	3.98**	4.04**
15	NN ₂ -2	36.00*	51.9*	41.0*	1.60	21.40**	4.85**	3.31**	3.41**
16	NN ₂ -3	36.70*	54.3*	41.0*	1.63	21.10**	4.83**	3.64**	3.71**
17	NN ₂ -7	40.50	57.9*	31.2*	1.77**	24.00**	4.80**	3.40**	4.09**
18	NN ₂ -8	45.50	62.4*	34.1*	1.77**	25.00**	4.46**	3.38**	4.36**
19	NN ₂ -10	45.50	62.4*	33.9*	1.70**	30.20**	4.06	3.50**	4.42**
20	NN ₂ -11	47.00**	63.0*	31.2*	1.80**	29.60**	4.22	3.29**	4.12**
21	NN ₂ -12	49.10**	66.3	45.1*	1.90**	26.20**	4.75**	3.67**	4.35**
22	NS ₂ N ₂ -3	48.40**	64.9*	40.3*	1.70**	28.90**	4.22	3.30**	3.90**
23	NS ₂ N ₂ -7	46.40	60.8*	32.9*	1.47	27.30**	4.43**	3.20**	3.96**
24	NS ₂ N ₂ -9	45.00	62.7*	34.9*	1.73**	27.20**	4.23	3.41**	4.01**
25	NE ₁ S ₁ -1	48.50**	66.3	34.7*	1.70**	27.80**	4.25	3.61**	4.19**
26	NE ₁ S ₁ -2	47.30**	62.3*	42.6*	1.67	30.00**	4.72**	3.26**	4.74**
27	NE ₁ S ₁ -3	46.20	62.7*	32.7*	1.50	26.20**	4.78**	3.75**	4.89**
28	NE ₁ S ₁ -4	45.20	63.3*	44.7*	1.67	24.00**	4.22	3.99**	4.13**
29	NE ₁ S ₁ -5	44.70	60.7*	41.3*	1.53	24.00**	4.60**	3.92**	4.35**
30	NE ₁ S ₁ -6	44.50	61.7*	49.0	1.63	23.00**	4.58**	3.70**	3.92**
31	NE ₁ S ₁ -7	41.20*	56.7*	30.9*	1.37*	24.60**	4.99**	3.16	4.20**
32	NE ₁ S ₁ -8	42.70*	58.7*	31.2*	1.63	28.30**	4.86**	3.38**	4.67**
33	NE ₁ S ₁ -9	42.30*	60.4*	41.1*	1.67	24.30**	4.22	4.07**	4.40**
34	NE ₁ S ₁ -10	40.50*	59.0*	42.0*	1.67	26.20**	4.67**	3.38**	4.03**
35	NE ₁ S ₁ -11	42.30*	60.6*	43.6*	1.80*	27.50**	4.65**	3.70**	4.30**
36	NE ₁ S ₁ -12	38.70*	54.7*	40.7*	1.37*	30.80**	4.05	3.84**	4.40**
37	NS ₁ N ₂ -1	38.70*	54.7*	38.3*	1.63	25.60**	4.80**	3.02	4.32**
38	NS ₁ N ₂ -3	39.20*	55.3*	37.5*	1.47	24.60**	4.65**	3.98**	3.80**
39	NS ₁ N ₂ -4	40.30*	58.9*	41.2*	1.70**	24.20**	4.33**	3.99**	4.13**
40	NS ₁ N ₂ -7	43.50	59.7	37.8*	1.70**	21.20**	4.69**	3.78**	3.66**
41	NS ₁ N ₂ -8	44.30*	60.4	37.4*	1.67	23.10**	4.91**	3.69**	3.90**
42	NS ₁ N ₂ -9	44.70	60.7*	37.3*	1.57	20.20**	4.72**	3.99**	3.90**
43	NS ₁ N ₂ -10	42.70*	58.7*	40.7*	1.67	24.00**	4.95**	3.51**	3.88**
44	NS ₁ N ₂ -11	41.30*	57.3*	36.7*	1.37	24.70**	4.36*	3.97**	4.24**
45	NS ₁ N ₂ -12	44.30*	61.5*	37.7*	1.77**	26.30**	4.62**	3.67**	4.25**
46	NS ₁ N ₂ -5	44.30*	60.5*	39.2*	1.60	22.70*	4.94**	3.90**	4.50**
47	Control	45.30	67.0	48.2	1.57	18.50	4.24	3.04	3.12
CD(0.05)		0.91	1.23	1.98	0.11	0.65	0.05	0.13	0.09

* Significant decrease at 0.05

** Significant increase at 0.05

over control.

Seeds/pod : The variation observed in seeds/pod of the mutant cultures ranged from 3.02 to 5.11 in Dhauri and 4.06 to 4.99 in Nayagarh local. The mean of this character observed in the controls plants of Dhauri and Nayagarh local was 4.51 and 4.24. Significant increase in seeds/pod over control was observed in twenty three and thirty one culture of Dhauri and Nayagarh local respectively.

100-seed weight : The range of variation in 100-seed weight (g) was from 3.09 to 5.08 and 3.01 to 4.07 in Dhauri and Nayagarh local respectively as against 3.27 and 3.04 in the controls. It was observed that twenty eight cultures of Dhauri and forty two from Nayagarh local recorded significant increase in this character over control. In Dhauri, ten mutant cultures recorded significant decrease in this trait as compared to control.

Yield/plant : The yield/plant (g) of the mutant cultures of Dhauri and Nayagarh local ranged from 3.86 to 5.29 and 3.41 to 4.67 as against 3.55 and 3.12 in the respective control. It was observed that all the mutant cultures of Dhauri and Nayagarh local recorded a significant increase in yield/plant over the control. The increase in yield ranged from 123.79 ($DE_1S_1 - 3$) to 170.09 ($DE_1S_1 - 5$) per cent in Dhauri and 105.49 ($NS_1 - 11$) to 151.28 ($NE_1S_1 - 2$) per cent in Nayagarh local in M_4 generation.

4.8.2 Variation, heritability and Genetic advance

Studies were undertaken to explore the possibilities of inducing micromutations in quantitative characters of mutant of

mutant cultures after chemical treatment. It was observed that there existed a wide range of variation for all eight characters studied in both the varieties. The genetic parameters like coefficient of variability, heritability and genetic advance of the quantitative traits were estimated and are presented in Tables 26 and 27.

The variation for different characters studied in terms of phenotypic coefficient of variability (PCV) and genotypic coefficient of variability (GCV) among the mutant cultures of Dhauri indicated that the PCV estimates were medium for plant height (19.13 %), pods/plant (21.69 %), and 100-seed weight (15.10 %). The PCV estimated were low for rest of the characters studied. Medium estimates of GCV were observed for plant height (18.92 %), pods/plant (21.61 %), and branches/plant (14.95 %). In case of Nayagarh local, PCV estimates were medium for plant height (12.87 %) and pods/plant (12.55 %). For rest of the characters this estimates were low. The GCV estimate was medium for plant height (12.44 %) and pods/plant (12.45 %). For rest of the characters the estimates were low. The PCV and GCV estimates of both the varieties, for all the eight characters studied followed almost similar trend. The PCV estimates were higher than the respective GCV estimates for all the characters studied.

The heritability estimates in broad sense for different characters varied from 41.71 % for 100-seed weight to 99.5 % for seeds per pod for the mutant cultures of Dhauri indicating the presence of high heritability. In the mutant cultures of Nayagarh local, it ranged from 80.8 % for branches/plant to

Table 26. Genetic parameters of quantitative traits in mutant cultures, Var - Dhauli.

Sl. No.	Characters name	Mean	Range	C.V.	P.C.V. (%)	G.C.V. (%)	h ² (%)	G.A. (5%)	G.A. as % of mean
1	50% Flowering (days)	38.77	32.40-47.67	2.24	10.38	10.13	95.30	7.90	20.30
2	Maturity (days)	55.75	48.00-65.00	2.33	8.27	7.93	92.00	8.74	15.84
3	Plant height (cm)	41.11	29.37-61.23	2.84	19.13	18.92	97.80	15.85	38.55
4	Branches/ Plant	1.43	1.13-1.83	5.16	15.82	14.95	89.40	0.42	37.17
5	Pods / Plant	30.44	21.67-48.94	1.91	21.69	21.61	99.20	13.50	44.35
6	Seeds / Pod	4.36	3.02-5.11	0.85	12.19	12.16	99.50	1.09	25.00
7	100-Seed weight (g)	3.66	3.02-4.96	11.52	15.10	9.75	41.71	0.47	12.84
8	Plant yield (g)	4.59	3.55-5.29	1.54	9.60	9.47	97.41	0.88	19.17

Table 27. Genetic parameters of quantitative traits in mutant cultures, Var - Nayagarh local.

Sl. No.	Characters name	Mean	Range	C.V.	P.C.V. (%)	G.C.V. (%)	h ² (%)	G.A. (5%)	G.A. as % of mean
1	50% Flowering (days)	44.02	38.67-50.67	1.29	7.95	7.84	97.4	7.02	15.95
2	Maturity (days)	60.76	51.98-66.33	1.31	5.89	5.74	95.1	7.00	11.52
3	Plant height (cm)	37.84	31.17-49.00	3.28	12.87	12.44	93.5	9.38	24.78
4	Branches/ Plant	1.64	1.37-1.90	4.09	9.33	8.39	80.8	0.25	15.24
5	Pods / Plant	25.39	18.48-30.82	1.61	12.55	12.45	98.4	6.46	25.44
6	Seeds / Pod	4.57	4.06-4.99	0.66	7.28	7.25	99.2	0.68	14.88
7	100-Seed weight (g)	3.56	3.04-4.07	2.32	8.67	8.25	92.8	0.59	16.57
8	Plant yield (g)	4.09	3.12-4.67	1.33	8.59	8.48	97.6	0.71	17.36

99.2 % for seeds/pod, also showed the presence of high heritability in all the characters. In both the varieties high heritability estimates was observed for seeds/pod.

The expected genetic advance of the mutant cultures of Dhauli under 5 % selection intensity was moderate for plant height (15.85 %) and pods/plant (13.5). It was low for rest of the characters. The expected genetic advance was moderate for plant height (9.38), while, it was low for rest of the characters for Nayagarh local. Genetic gain when expressed as percentage of mean was moderate for pods/plant (25.44) and plant height (24.78) while, it was less for rest of the characters in Nayagarh local. The expected genetic advance when expressed as percentage of mean was high for pods/plant in both the varieties. However, high genetic advance coupled with high heritability was observed for pods/plant in both the varieties.

4.8.3 Interrelationship among characters

Inability to recognise small difference in quantitative traits among single plants had led to frequent attempts to find associated traits more amenable to visual selection. The genotypic (r_g) and phenotypic (r_p) correlation coefficients provides a measure of relationship between the traits and serve to assess the chance for mutual improvement of the two traits by a common selection or using selection for one trait to identify genotypes with superior expression of related traits. The correlation coefficient among eight quantitative characters of the mutant cultures of both the varieties were estimated and presented in Tables 28 and 29.

The phenotypic and genotypic correlation coefficient

estimates in the mutant cultures of Dhauli revealed that the inter relationship between yield and branches/plant was highly significant and positive both at genotypic and phenotypic level. The correlation coefficient between yield and pods/plant and 100-seed weight were significant and positive at genotypic level. The association between yield and 50 % flowering and maturity was significant but negative at 1 % level. The association between yield and rest of the characters was not significant.

In the cultures of Dhauli, the correlation coefficient at genotypic and phenotypic level indicated that days to 50 % flowering was highly significant and positive with days to maturity. The genotypic and phenotypic correlations of branches/plant with days to 50 % flowering and maturity was negative but highly significant. The correlation of pods/plant with other characters, was positive but not significant for majority of the cases. Seeds/pod had a negative correlation with majority of characters and it's negative association with plant height and pods/plant was significant. The correlation coefficient between 100-seed weight and pods/plant was significant in a negative direction, but its association with seeds/pod was positive and significant.

The correlation coefficient between yield and pods/plant was significant and positive both at genotypic and phenotypic level in case of Nayagarh local. The correlation coefficient estimates between yield and rest of the characters was not significant, though some were positive and others were negative.

Table 28. Correlation, phenotypic (r_p) and genotypic (r_g) among the quantitative traits of mutant cultures, Var - Dhuli.

Sl. No.	Characters Name	Correlations	Days to maturity	Plant height	Branches/ Plant	Pods/ Plant	Seeds/ Pod	100-Seed weight	Plant yield
1	50% flowering (days)	r_p	0.777**	0.063	-0.556**	0.005	-0.264	-0.186	-0.614**
		r_g	0.823**	0.063	-0.612**	0.012	-0.267	-0.291*	-0.640**
2	Maturity (days)	r_p		0.199	-0.487**	-0.085	-0.202	-0.066	-0.522**
		r_g		0.209	-0.541**	-0.095	-0.213	-0.046	-0.557**
3	Plant height	r_p			-0.009	0.217	-0.353*	-0.004	-0.057
		r_g			-0.001	0.220	-0.359*	-0.003	-0.058
4	Branches/ Plant	r_p				0.129	0.131	0.105	0.609**
		r_g				0.139	0.141	0.347*	0.662**
5	Pods/ Plant	r_p					-0.775**	-0.297*	0.297
		r_g					-0.782**	-0.463**	0.301*
6	Seeds/ Pod	r_p						0.271	0.134
		r_g						0.420**	0.135
7	100-Seed weight	r_p							0.219
		r_g							0.348*

* Significant at 0.05

** Significant at 0.01

Table 29. Correlation, phenotypic (r_p) and genotypic (r_g) among the quantitative traits of mutant cultures, Var - Nayagarh local.

Sl. No.	Characters	Correlations	Days to maturity	Plant height	Branches/ Plant	Pods/ Plant	Seeds/ Pod	100-Seed weight	Plant yield
1	50% flowering (days)	r_p	0.884**	-0.220	0.355*	0.084	-0.078	0.01	0.136
		r_g	0.866**	-0.211	0.312*	0.081	-0.070	0.013	0.135
2	Maturity (days)	r_p		-0.054	0.447**	0.019	-0.181	-0.015	0.047
		r_g		-0.053	0.389**	0.014	-0.177	-0.011	0.047
3	Plant height	r_p			-0.115	-0.014	-0.152	0.060	-0.081
		r_g			-0.101	0.014	-0.147	0.057	-0.081
4	Branches/ Plant	r_p				-0.211	0.120	0.091	-0.145
		r_g				-0.188	0.144	0.067	-0.128
5	Pods/ Plant	r_p					-0.303*	-0.374*	0.609**
		r_g					-0.298*	-0.357*	0.619**
6	Seeds/ Pod	r_p						-0.093	0.021
		r_g						-0.090	0.021
7	100-Seed weight	r_p							0.136
		r_g							0.129

* Significant at 0.05

** Significant at 0.01

In case of Nayagarh local, correlation between 50 % flowering and maturity was highly significant in a positive direction. Branches/plant was positive and significantly correlated with days to 50 % flowering and maturity. None of the correlation coefficients, between pods/plant and other characters was found significant. Seeds/pod was negatively correlated with all other characters. The correlation coefficient at genotypic and phenotypic level between 100-seed weight and pods/plant were highly significant in a negative direction.

4.8.4 Path coefficients

The relationship between the traits expressed by correlation coefficient (r) of the cultures of both the varieties was partitioned into direct and indirect contributions of component traits towards the expression of yield are presented in Tables 30 and 31. At genotypic level, highest direct effect was expressed by pods/plant (0.960) followed by seeds/pod (0.607) and 100-seed weight (0.410) for the mutant cultures of Dhauli. The negative direct effect was expressed by the days to 50 % flowering, days to maturity, and plant height. The total genotypic correlation of days to 50 % flowering and days to maturity with yield was negative and significant at 1 % level, due to its negative direct effect and negative indirect effect via rest of the characters. Though the strength of the direct effect of branches/plant was low, the genotypic correlation with yield was enhanced due to positive indirect effect via all other characters. On the contrary, the genotypic correlation coefficient between

Table 30. Direct and indirect effect of component traits on seed yield at genotypic level of mutant cultures of Dhauri.

Sl. No.	Character	Days to 50% Flowering	Days to maturity	Plant height (cm)	Branches/ Plant	Pods/ Plant	Seeds/ Pod	100-Seeds weight (g)	Total genotypic correlation with yield
1	Days to 50% Flowering	<u>-0.290</u>	-0.006	-0.002	-0.072	0.011	-0.162	-0.119	-0.640**
2	Days to Maturity	-0.239	<u>-0.008</u>	-0.007	-0.064	-0.091	-0.129	-0.019	-0.557**
3	Plant height (cm)	-0.018	-0.002	<u>-0.031</u>	0.001	0.211	-0.218	0.001	-0.058
4	Branches/ Plant	0.178	0.004	0.001	<u>0.117</u>	0.134	0.086	0.142	0.662**
5	Pods/ Plant	-0.003	0.001	-0.007	0.016	<u>0.960</u>	-0.475	-0.190	0.301*
6	Seeds/ Pod	0.077	0.002	0.011	0.017	-0.751	<u>0.607</u>	0.172	0.135
7	100-Seeds weight (g)	0.084	0.001	0.001	0.041	-0.444	0.255	<u>0.410</u>	0.348*

Residual effect = 0.2165
 Figures underlined indicate direct effect.

Table 31. Direct and indirect effect of component traits on seed yield at genotypic level of mutant cultures of Nayagarh local.

Sl. No.	Character	Days to 50% Flowering	Days to maturity	Plant height (cm)	Branches/ Plant	Pods/ Plant	Seeds/ Pod	100-Seeds weight (g)	Total genotypic correlation with yield
1	Days to 50% Flowering	<u>-0.145</u>	0.26	0.016	-0.049	0.076	-0.028	0.005	0.135
2	Days to Maturity	-0.128	<u>0.294</u>	0.004	-0.061	0.017	-0.071	-0.008	0.047
3	Plant height (cm)	0.032	-0.016	<u>-0.073</u>	0.016	-0.013	-0.059	0.032	-0.081
4	Branches/ Plant	-0.051	0.131	0.008	<u>-0.138</u>	-0.101	0.047	0.048	-0.128
5	Pods/ Plant	-0.012	0.006	0.001	0.029	<u>0.903</u>	-0.119	-0.199	0.609**
6	Seeds/ Pod	0.011	-0.053	0.011	-0.016	-0.274	<u>0.391</u>	-0.049	0.021
7	100Seeds weight (g)	-0.001	-0.004	-0.004	-0.012	-0.345	-0.036	<u>0.531</u>	0.129

Residual effect = 0.3508
 Figures underlined indicate direct effect.

Pods/plant and yield, was lowered due to negative indirect effect via days to flowering, and seeds/pod even though its direct effect was highest. The direct effect of seeds/pod though high, it was nullified due to high indirect effect via pods/plant causing a low genotypic correlation with yield.

In the mutant cultures of Nayagarh local, highest direct effect was observed by pods/plant (0.903), followed by 100-seed weight (0.531), seeds/pod (0.391) and days to maturity (0.294). The direct effect was negative for days to flowering, plant height and branches/plant. The total genotypic correlation of 50 % flowering with yield could become positive, nullifying its negative direct effect, due to positive indirect effect via days to maturity, plant height, pods/plant, and 100-seed weight. The total genotypic correlation between 100-seed weight and yield was reduced, due to negative indirect effect via all the characters, even though its direct effect was high. Similarly, though the direct effect of seeds/pod was high, it was nullified due to high indirect effect via pods/plant, days to maturity, resulting a lower genotypic correlation with yield.

4.8.5 GENETIC DIVERGENCE

The differences among the 47 cultures including parent derived from Dhauli and Nayagarh local by mutagenic treatment were assessed by estimating the genetic diversity through multivariate analysis of D^2 statistics and presented here with.

Based on the degree of divergence 47 mutant cultures of Dhauli and Nayagarh local were grouped into 11 and 12 clusters respectively using D^2 value in such a way that the cultures

Table 32. Clustering pattern of mutant cultures of Dhauli on the basis of genotypic divergence.

Cluster No.	Mutant clusters included	No. of genotypes
I	DE ₁ - 4, DN ₂ - 5, DE ₁ S ₁ - 4, DE ₁ S ₁ - 9, DE ₁ S ₁ -10, DS ₁ N ₂ - 6, DS ₁ N ₂ -11	7
II	DE ₂ - 11, DE ₁ S ₁ - 3, DC	3
III	DE ₁ S ₁ - 4, DS ₁ N ₂ - 7, DS ₁ N ₂ - 8, DS ₁ N ₂ - 9	4
IV	DE ₁ - 3	1
V	DE ₁ S ₁ - 5, DE ₁ S ₁ - 7, DE ₂ N ₁ - 2	3
VI	DE ₂ - 6, DE ₂ - 8, DE ₂ - 9, DE ₂ -12, DN ₂ -1, DE ₂ N ₁ - 4	6
VII	DE ₁ -12, DN ₂ - 5	2
VIII	DN ₂ - 7, DN ₂ - 8, DE ₁ S ₁ -1	3
IX	DE ₁ S ₁ -11, DE ₁ S ₁ -12, DS ₁ N ₂ -1, DS ₁ N ₂ -2, DS ₁ N ₂ - 3, DS ₁ N ₂ - 4, DS ₁ N ₂ - 5	7
X	DE ₁ -1, DE ₁ -10, DE ₂ -1, DE ₂ - 3, DE ₂ - 4, DE ₂ N ₁ - 5, DE ₂ N ₁ - 8, DE ₂ N ₁ -11,	9
XI	DS ₁ N ₂ -10, DS ₁ N ₂ -12	2

Table 33. Clustering pattern of mutant cultures of Nayagarh local on the basis of genotypic divergence.

Cluster No.	Mutant clusters included	No. of genotypes
I	NN ₂ -2, NN ₂ -3, NS ₁ N ₂ -3	3
II	NS ₁ -7, NS ₁ -11	2
III	NS ₂ - 4, NE ₁ S ₁ - 7, NE ₁ S ₁ - 8	3
IV	NE ₁ - 3, NS ₂ - 9, NN ₂ - 8, NN ₂ -10, NN ₂ -11, NS ₂ N ₂ - 7, NS ₂ N ₂ - 9, NE ₁ S ₁ -1	8
V	NE ₁ S ₁ - 4, NE ₁ S ₁ - 6, NE ₁ S ₁ -9, NE ₁ S ₁ -11, NS ₁ N ₂ - 4, NS ₁ N ₂ -12	6
VI	NC	1
VII	NE ₁ - 6, NE ₁ -10, NE ₁ - 8, NE ₁ S ₁ -12, NS ₁ N ₂ -11	5
VIII	NS ₂ -10, NS ₂ N ₂ -3, NE ₁ S ₁ -2	3
IX	NE ₁ -11, NN ₂ -7, NE ₁ S ₁ -10, NE ₁ N ₂ -1, NE ₁ N ₂ -10	5
X	NS ₂ -2, NS ₂ -12, NS ₁ N ₂ -7, NS ₁ N ₂ - 8, NS ₁ N ₂ -9	5
XI	NE ₁ S ₁ - 3, NE ₁ S ₁ - 5, NS ₁ N ₂ - 5	3
XII	NE ₁ - 5, NS ₁ - 6, NN ₂ -12	3

Table 34. Average of Intra and Inter Cluster distance (D^2) values for the Dhauili.

Cluster No.	I	II	III	IV	V	VI	VII	VIII	IX	X	XI
I	1.532 (1.248)	13.03 (3.61)	6.60 (2.47)	29.70 (5.45)	7.95 (2.82)	11.49 (3.39)	14.89 (3.86)	23.04 (4.80)	2.31 (1.52)	10.17 (3.19)	6.35 (2.52)
II		1.769 (3.09)	(4.69)	(4.49)	(5.34)	(3.50)	(3.61)	(4.33)	(4.25)	(2.93)	(5.48)
III			1.118 (1.16)	(4.56)	(3.80)	(4.19)	(3.00)	(4.16)	(2.86)	(4.56)	(2.03)
IV				0.000 0.0	(6.72)	(3.38)	(3.25)	(3.65)	(5.75)	(5.00)	(5.44)
V					45.15	15.79	10.56	13.32	33.06	25.00	29.50
VI					(1.69)	(4.21)	(5.00)	(4.79)	(1.53)	(4.01)	(3.51)
VII					2.85	16.79	25.00	22.94	2.35	16.81	12.32
VIII						(1.02)	(3.15)	(2.73)	(4.04)	(2.16)	(4.51)
IX						1.04	9.92	7.45	16.32	4.66	20.34
X							(1.18)	(2.87)	(3.89)	(3.90)	(4.10)
XI							1.32	8.23	15.13	15.21	16.81
								(1.13)	(5.29)	(4.61)	(4.68)
								1.27	27.98	21.25	21.90
									(1.34)	(3.60)	(2.60)
									1.74	12.96	6.76
										(1.20)	(4.29)
										1.44	18.40
											(0.48)
											0.23

* Figure in the parenthesis denotes 'D' value.

Table 35. Average of Intra and Inter Cluster distance (D^2) values for the Nayagarh local.

Cluster No.	I	II	III	IV	V	VI	VII	VIII	IX	X	XI	XII
I	1.860 (1.364)	25.270 (5.027)	14.900 (3.860)	(23.02) (4.798)	13.003 (3.606)	23.415 (4.839)	14.280 (3.779)	27.321 (5.227)	5.958 (2.441)	15.276 (3.908)	17.347 (4.165)	32.913 (5.737)
II		1.485 (1.219)	29.278 (5.411)	14.455 (3.802)	17.740 (4.212)	31.225 (5.588)	34.047 (5.835)	5.914 (2.432)	19.236 (4.386)	5.942 (2.437)	23.716 (4.870)	11.405 (3.361)
III			3.697 (1.923)	12.659 (3.556)	16.941 (4.116)	44.023 (6.635)	13.191 (3.632)	14.853 (3.854)	6.492 (2.548)	13.249 (3.640)	11.075 (3.328)	17.089 (4.134)
IV				1.638 (1.280)	8.031 (2.834)	36.030 (6.050)	9.628 (3.103)	5.112 (2.261)	8.392 (2.897)	8.661 (2.943)	9.265 (3.044)	5.424 (2.329)
V					2.067 (1.438)	36.240 (6.020)	9.036 (3.006)	8.874 (2.979)	5.982 (2.466)	5.271 (2.296)	4.309 (2.076)	9.696 (3.114)
VI						(0.000) 0.000	34.368 (5.864)	24.750 (4.975)	33.640 (5.800)	26.204 (5.119)	41.602 (6.450)	40.972 (6.401)
VII							2.509 (1.584)	10.252 (3.202)	8.614 (2.935)	14.600 (3.821)	8.450 (2.907)	21.659 (4.654)
VIII								2.137 (1.462)	10.214 (3.196)	16.329 (4.041)	11.089 (3.330)	7.901 (2.811)
IX									2.715 (1.648)	8.305 (2.882)	9.966 (3.157)	15.272 (3.908)
X										1.157 (1.076)	5.085 (2.255)	8.803 (2.967)
											1.012 (1.006)	8.673 (2.945)
												2.611 (1.616)

* Figure in the parenthesis denotes 'D' value.

within a cluster had smaller D^2 values than those in other clusters. Clustering pattern of cultures was based on the degree of divergence between the cultures. Composition of different clusters with their corresponding cultures included in each clusters have been given in Tables 32 and 33. ^(Fig 3,4) It was revealed from the tables that cultures derived from any of the mutagens either single or in combination fell into same cluster. Further, it was observed that cluster IV, ^{and} VII of Dhauri and cluster II and XII of Nayagarh local were comprised of cultures derived from single mutagenic treatment only. In the same way, cluster III, IX and XI of Dhauri and cluster V, XI of Nayagarh local included cultures derived from combined treatment of mutagens only. The rest clusters in both the sets of experiment included cultures derived both from single and combined mutagenic treatment.

Canonical analysis was done for confirmation of clustering pattern of mutant cultures and are presented in Figures 1 and 2. The first two canonical root in the study accounted 50.03 (Z_1) and 19.18 (Z_2) for Nayagarh local and 74.38 (Z_1) and 12.79 (Z_2) for Dhauri percentage of diversity among the cultures. The position of cultures in the figure corroborated the grouping done by Tocher's method.

Intra and inter cluster distance

The difference between the mutant cultures of both the varieties was assessed by estimating genetic diversity and presented herewith (Tables 35 and 36). (Figure 1 and 2) It was revealed that the intra cluster distance varied from 0.23 to 3.09 in Dhauri, cluster II was having the highest intra cluster

distance of 3.09. It was indicated that maximum genetic diversity (3.09) was present among the three cultures of that group. It was followed by cluster IV (2.85) having similar numbers of mutant cultures. Cluster IV had no intra - cluster distance as it was represented by a single culture. The maximum genetic distance (45.15) was depicted between the cluster V and VI followed by between cluster IV and IX (33.06). Cluster IX revealed the least divergence relationship with cluster I (2.31).

With regards to the intra-cluster distance among the cultures of Nayagarh local, it was revealed that the range of genetic distance (D^2) was from 0.0 to 3.697. The maximum value of intra-cluster distance was noticed in cluster-III, which included only three cultures in it. Nevertheless cluster VI had no intra cluster distance as it was represented by a single culture that to the parent variety. As regards to inter cluster distance, highest divergence ($D^2 = 44.023$) was noticed between cluster VI and III, followed by between XI and VI (41.602) and between XII and VI (40.972). The lowest inter-cluster distance was observed between cluster XI and V (4.309). The genetic distance was moderate between rest of the clusters except between cluster I and IX, between cluster III and IX, cluster IV and IX, cluster V and IX having D^2 values just around 2.0.

Cluster means for different characters

The mean performances of eight different characters of eleven clusters of Dhauli and twelve clusters of Nayagarh local was studied and presented in Tables 37 and 38 respectively. It was revealed from the means of the cultures of Dhauli that

there was marked variation between clusters for plant height, pods/plant and grain yield/plant. Cluster IV had the low mean value for plant height (28.53 cm) indicating the presence of dwarf plant types in it whereas cluster V (54.41 cm) and VIII (55.06 cm) had tall plants in it. Semi tall plants were observed in rest of the clusters, the means of which varied from 34.98 cm to 45.47 cm. The mean for days to maturity varied from 50.45 (cluster XI) to 61.24 (cluster X) indicating the presence of early mutant cultures in the former cluster. Mean pods/plant was highest in cluster IV (48.94) followed by cluster VIII (43.17) and cluster VII (40.86). The mean number of pods/plant was lowest in cluster II (24.86). Highest and lowest mean number of seeds/pod was observed in cluster IX (4.76) and cluster IV (3.05). With respect to yield/plant, Cluster III occupied the top position (5.16 g/plant) followed by cluster XI (5.10). This value was lowest in cluster II (3.75 g). The clusters of top yielding clusters were having short height associated with high pods/plant, seeds/pod and moderate 100-seed weight. Cultures in cluster IV had the highest pods/plant but owing to low 100-seed weight and seeds/pod, the yield/plant was comparatively reduced.

The different cluster of Nayagarh local indicated a marked variation in the means for duration, plant height and pods/plant. Late maturing cultures were present in cluster VI, XII whereas medium duration types were found in rest of the clusters. The variation in mean height among the cultures were from 31.43 cm (cluster II) to 48.23 (cluster VI). The mutant cultures having intermediate plant height were grouped in rest

Table 36. Group mean of eight character in different clusters , Var - Dhauli.

Cluster No.	50% Flowering (days)	Maturity (days)	Plant height (cm)	Branches/ Plant	Pods/ Plants	Seeds/ Pod	100-seed weight (g)	Yield/ Plant (g)
I	36.70	52.68	38.53	1.64	26.54	4.67	3.81	4.62
II	39.42	56.46	44.19	1.27	24.86	4.43	2.91	3.75
III	35.48	52.95	41.67	1.77	37.37	4.36	3.37	5.16
IV	42.00	59.33	28.53	1.43	48.94	3.05	3.09	4.63
V	37.03	55.93	54.41	1.54	27.10	4.73	4.52	4.19
VI	42.06	59.21	45.47	1.28	30.18	3.85	3.62	4.17
VII	40.83	52.67	42.70	1.25	40.86	4.06	2.91	4.87
VIII	40.56	58.08	55.06	1.33	43.17	3.26	3.42	4.48
IX	34.19	50.94	34.98	1.42	27.17	4.76	3.89	4.93 4.99
X	42.99	61.26	36.51	1.23	26.19	4.70	3.71	4.36
XI	34.33	50.45	37.42	1.75	36.27	4.01	4.12	5.10

Table 37. Group mean of eight character in different clusters , Var - Nayagarh local.

Cluster No.	50% Flowering (days)	Maturity (days)	Plant height (cm)	Branches/ Plant	Pods/ Plants	Seeds/ Pod	100-seed weight (g)	Yield/ Plant (g)
I	37.28	53.83	39.84	1.57	22.38	4.77	3.64	3.64
II	46.83	63.83	31.43	1.88	19.64	4.60	3.67	3.38
III	41.74	58.20	33.59	1.54	27.09	5.22	3.18	4.32
IV	46.24	63.13	33.33	1.68	27.37	4.32	3.42	4.14
V	43.17	61.07	42.86	1.71	24.88	4.44	3.85	4.19
VI	45.33	66.93	48.23	1.57	18.48	4.24	3.04	3.12
VII	42.41	58.02	38.97	1.37	28.26	4.25	3.59	4.22
VIII	46.71	63.70	42.62	1.63	29.71	4.42	3.20	4.23
IX	40.27	57.29	38.65	1.70	25.64	4.65	3.35	4.04
X	45.23	60.88	35.81	1.67	21.85	4.81	3.88	3.93
XI	45.06	61.29	39.40	1.54	24.34	4.77	3.86	4.58
XII	49.66	65.96	36.83	1.80	26.14	4.78	3.56	4.32

of the cultures except culture V and VIII. The highest mean number of pods/plant was recorded in cluster VIII (29.71) followed by cluster VII (28.26) and cluster IV (27.37) whereas lowest in cluster II (19.64), Seeds/pod varied from 4.24 (cluster VI) to 5.22 in cluster III. Mean yield/plant was the highest in cluster XI (4.58 g/plant) and lowest in cluster VI (3.12 g/plant).

Relative contribution of characters to genetic divergence

The relative contribution of different characters to the total divergence was calculated by rank average method and presented in Table 39. In cases where more number of characters occupied first rank, the rank total in that case obviously becomes low indicating more contribution of the character to the total divergence. In Dhauri, it was revealed from the data that 100-seed weight contributed the maximum towards genetic divergence. The contribution of other characters to the divergence were in order of days to maturity, branches/plant, days to 50% flowering, plant height, yield/plant, pods/plant and seeds/pod. The relative contributions of branches/plant to the genetic divergence was highest in case of Nayagarh local. It was followed by days to maturity and plant height.

Considering the contributions of different characters to the total divergence, it was observed that different characters in both the varieties contributed differently to the diversity. However, in the present investigation contribution of seeds/pod and pods/plant to the total divergence was low in both the sets of experiments.

Table 38. Relative contribution of different characters to genetic diversity among different mutant cultures of Dhauli and Nayagarh local.

Sl. No.	Characters	Dhuli		Nayagarh local	
		Rank total	Percentage of Rank total	Rank total	Percentage of Rank total
1	Days to 50% Flowering	5227	13.431 (4)	4239	10.180 (5)
2	Maturity (days)	6294	16.173 (2)	6330	16.266 (2)
3	Plant height (cm)	4528	11.635 (5)	5463	14.038 (3)
4	Branches/Plant	5721	14.709 (3)	6617	17.004 (1)
5	Pods/Plant	3498	8.988 (7)	3571	9.181 (7)
6	Seeds/Pod	2833	7.279 (8)	3017	7.752 (8)
7	100-Seed weight (g)	6908	17.751 (1)	5447	13.997 (4)
8	Yield/Plant (g)	3907	10.039 (6)	4220	10.844 (6)

Note :- Number in the parenthesis indicates order of Contribution (descending) to genetic diversity.

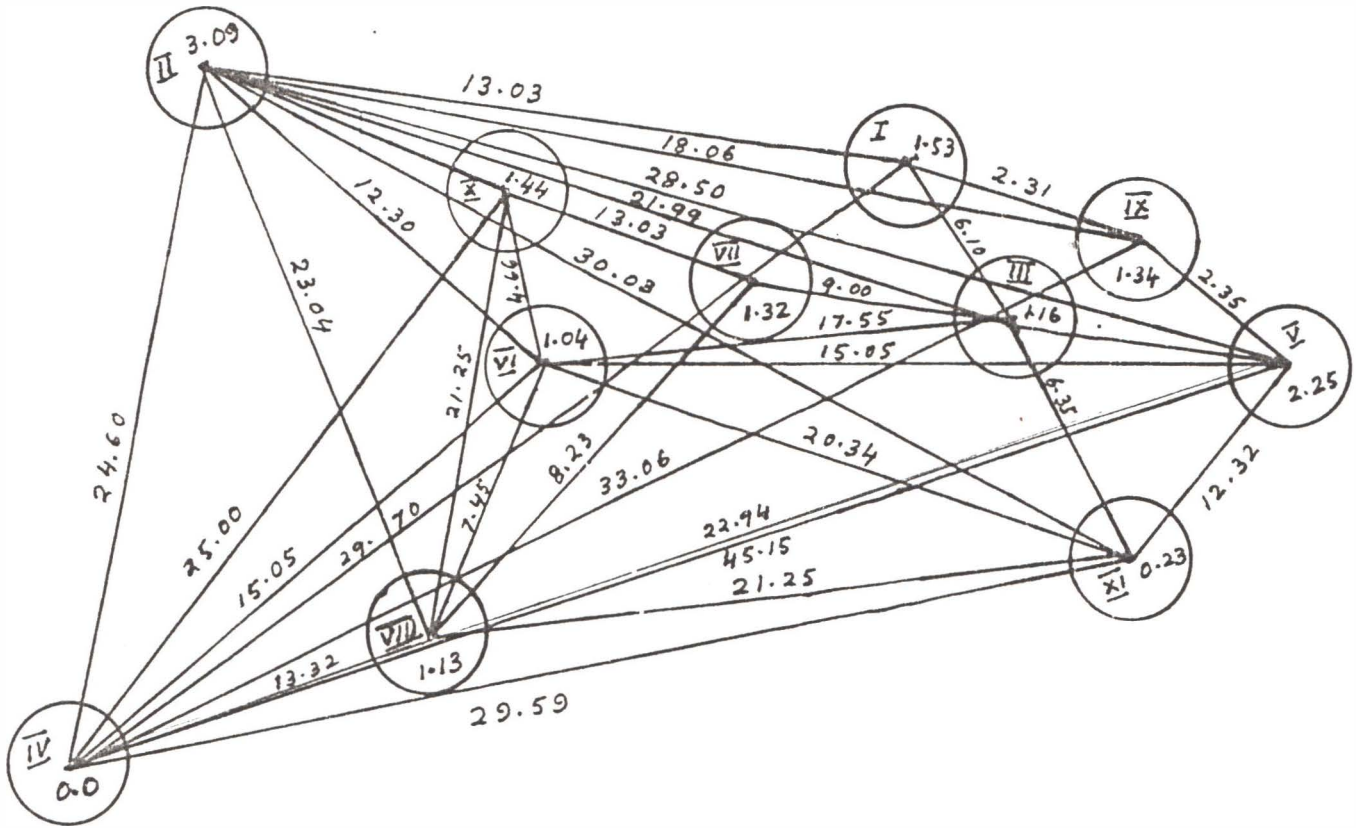


Fig.1. Statistical distance among 47 mutant lines of Dhaul1

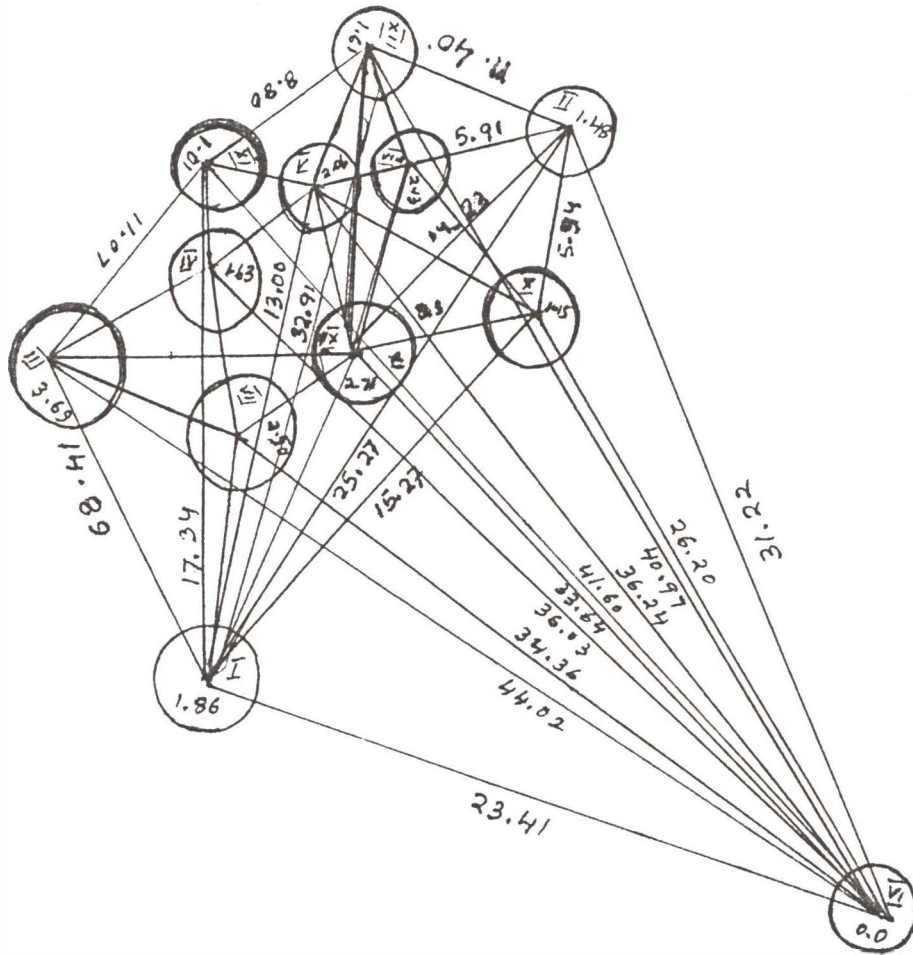


Fig. 2. Statistical distance among 47 mutant lines of Nayagarh local

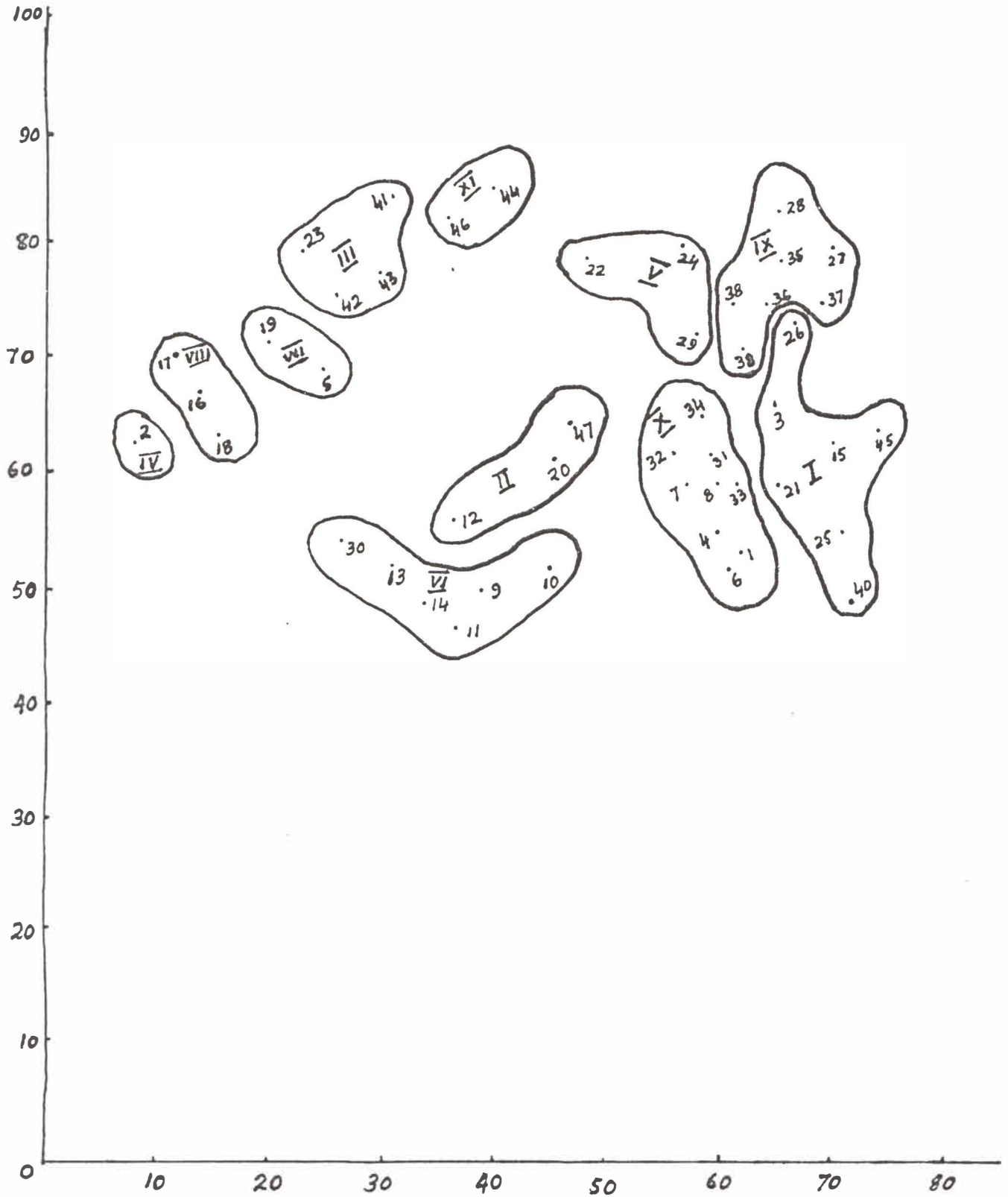


Fig.3. Scatter of points representing Dhauri in (Z_1-Z_2) graph with D^2 clustering super imposed

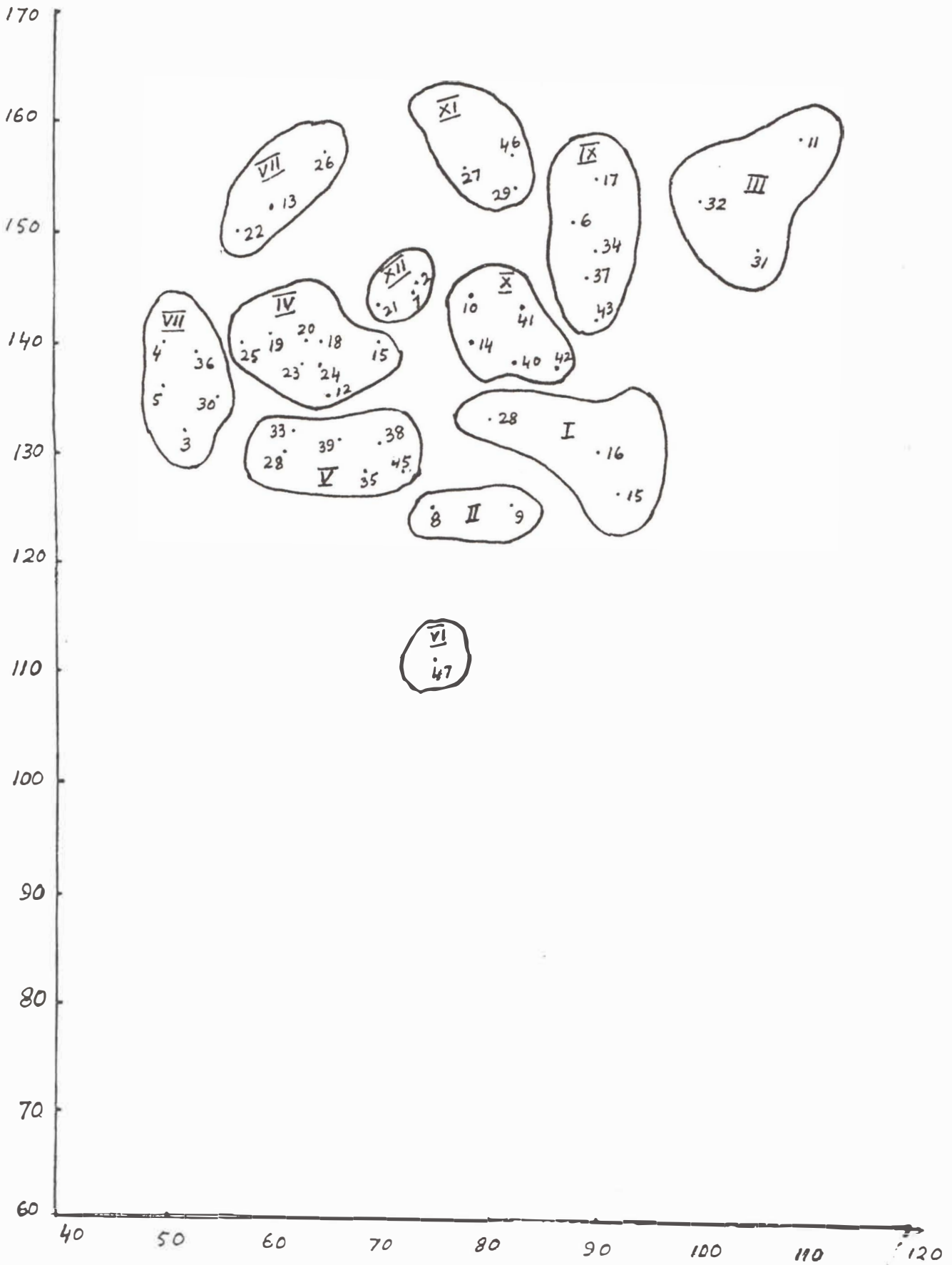


Fig.4. Scatter of points representing Nayagarh local in (Z_1-Z_2) graph with D^2 clustering super imposed

4.8.6 SELECTION INDICES

Yield is a complex character and is more often influenced by the environment for the expression of the trait. Therefore, selection based on the yield only is less effective. In the present study mutant cultures were selected both by direct method on yield alone and methods based on multiple criteria selection indices. Five different selection criteria (SC) were applied for selecting better yielding cultures. They were as follows.

SC - I. Direct selection based on yield/plant.

SC - II. Selection based on pods/plant, seeds/pod and 100-seed weight.

SC - III. Selection based upon pods/plant, seeds/pod, 100-seed weight and yield/plant.

SC - IV. Selection based on morphological traits like days to flowering, days to maturity, plant height and branches/plant.

SC - V. Selection based on the basis of all the 8 characters.

The discriminant functions were calculated for getting the index values of each cultures for different selection criteria and were presented below.

For Dhauli.

$$\text{SC - I.} = 0.9742 X_8$$

$$\text{SC - II.} = -2.1330 X_5 + 6.1903 X_6 + -11.3238 X_7$$

$$\text{SC - III.} = 0.009 X_5 + 0.4269 X_6 + -1.292 X_7 + - 0.5278 X_8$$

$$\text{SC - IV.} = -0.0392 X_1 + -0.0053 X_2 + -0.0014 X_3 + 0.7558 X_4$$

$$\text{SC - V.} = 0.001 X_1 + 0.001 X_2 + 0.001 X_3 + 0.004 X_4 + \\ 0.10 X_5 + 0.03 X_6 + 0.02 X_7 + 0.80 X_8$$

For Nayagarh local.

$$SC - I. = 0.9758 X_8$$

$$SC - II. = 0.7154 X_5 + -0.2878 X_6 + 0.0164 X_7$$

$$SC - III. = 0.7335 X_5 + -0.3153 X_6 + -0.1454 X_7 + -0.1055 X_8$$

$$SC - IV. = 0.038 X_1 + -0.0178 X_2 + -0.0018 X_3 + -0.3904 X_4$$

$$SC - V. = 0.001 X_1 + 0.001 X_2 + 0.001 X_3 + -0.002 X_4 + \\ 0.10 X_5 + 0.04 X_6 + 0.06 X_7 + 0.82 X_8$$

Where

X_1 = Days to flowering

X_5 = Pods/plant

X_2 = Days to maturity

X_6 = Seeds/pod

X_3 = Plant height

X_7 = 100-seed weight

X_4 = Branches/plant

X_8 = Yield/plant

These functions were used and index values for the cultures were estimated which are presented in Table 39 for Dhauri and Table 40 for Nayagarh local. On the basis of each selection criteria 9 out of 47 mutant cultures (20% selection intensity) were selected. Those selected cultures were ranked on the basis of five different criterion and are presented in Table 41. From the mutant cultures of Dhauri culture developed for E_1S_1 mutagenic treatment ranked first from Sc - I. Whereas, cultures of Nayagarh local developed from E_1S_1 ranked first each from Sc - I, SC - II and SC - III. One culture each of Dhauri (DE_1S_1) and Nayagarh local (NE_1S_1) were found positions in three selection criterias out of five whereas, rest ranked differently by different criteria.

Expected genetic gain for different selection criteria at 20% selection intensity were estimated and presented in Table

Table 39. Selection index values of mutant cultures of Dhauli on the basis of different selection criteria.

Sl. No.	Treatment Code	S.C - I	S.C - II	S.C - III	S.C - IV	S.C - V
1	NE ₁ -3	3.960	-68.315	-4.609	-1.234	6.197
2	NE ₁ -5	4.510	-120.494	-4.683	-0.920	8.886 (1)
3	NE ₁ -6	4.442	-68.191	-5.242	-0.618	6.478
4	NE ₁ -8	4.461	-72.145	-4.905	-1.222	6.822
5	NE ₁ -10	4.666	-92.158	-4.050 (3)	-1.009	8.151 (9)
6	NE ₁ -11	4.023	-68.036	-4.762	-1.375	6.231
7	NS ₁ -6	4.247	-60.746 (3)	-4.921	-1.201	6.103
8	NS ₁ -7	4.159	-73.031	-5.127	-1.056	6.437
9	NS ₁ -11	4.169	-80.503	-5.337	-1.149	6.632
10	NS ₂ -2	3.964	-70.331	-4.980	-0.970	6.093
11	NS ₂ -4	4.033	-84.032	-4.981	-0.945	6.780
12	NS ₂ -9	3.370	-70.392	-3.429 (1)	-0.895	6.485
13	NS ₂ -10	4.062	-83.566	-4.983	-1.078	6.745
14	NS ₂ -12	4.159	-79.852	-4.688	-1.232	6.848
15	NN ₂ -2	4.072	-62.208 (4)	-4.637	-0.794	6.081
16	NN ₂ -3	4.101	-98.928	-4.177 (4)	-0.897	7.813
17	NN ₂ -7	4.491	-118.567	-5.376	-0.962	8.535 (2)
18	NN ₂ -8	4.491	-114.265	-5.407	-1.047	8.322 (5)
19	NN ₂ -10	4.822	-97.902	-4.400 (6)	-0.980	8.455 (4)
20	NN ₂ -11	3.750	-50.044 (1)	-3.464 (2)	-0.791	5.625
21	NN ₂ -12	4.354	-70.145	-4.735	-0.479	6.686
22	NS ₂ N ₂ -3	5.153 (1)	-66.522 (8)	-4.232 (5)	-0.465 (6)	6.754
23	NS ₂ N ₂ -7	5.124 (3)	-67.009 (9)	-5.084	-0.512	8.461 (3)
24	NS ₂ N ₂ -9	5.114 (2)	-65.865 (7)	-5.783	-0.539	7.146
25	NE ₁ S ₁ -1	4.081	-78.236	-5.091	-0.564	6.564
26	NE ₁ S ₁ -2	4.539	-75.861	-4.789	-0.472 (8)	6.681
27	NE ₁ S ₁ -3	4.948 (9)	-68.585	-5.115	-0.478 (9)	7.090
28	NE ₁ S ₁ -4	4.958 (8)	-77.236	-5.219	-0.518	7.293
29	NE ₁ S ₁ -5	4.091	-88.585	-6.494	-0.861	6.578
30	NE ₁ S ₁ -6	3.984 (7)	-91.148	-4.759	-1.043	7.055
31	NE ₁ S ₁ -7	4.150	-64.805 (5)	-4.736	-0.973	6.229
32	NE ₁ S ₁ -8	4.530	-77.306	-4.859	-1.086	7.121
33	NE ₁ S ₁ -9	4.179	-68.844	-4.660	-1.067	6.468
34	NE ₁ S ₁ -10	4.530	-65.792 (6)	-4.880	-1.075	6.614
35	NE ₁ S ₁ -11	4.958	-74.589	-5.587	-0.441 (5)	7.173
36	NE ₁ S ₁ -12	4.900	-73.937	-5.942	-0.465 (7)	6.923
37	NS ₁ N ₂ -1	4.539	-69.842	-5.251	-0.642	6.651
38	NS ₁ N ₂ -3	4.617	-73.542	-5.265	-0.740	6.870
39	NS ₁ N ₂ -4	4.705	-72.782	-4.959	-0.840	7.057
40	NS ₁ N ₂ -7	4.568	-81.120	-5.241	-0.476	7.123
41	NS ₁ N ₂ -8	5.065 (5)	-83.202	-4.456 (8)	-0.350 (3)	7.963
42	NS ₁ N ₂ -9	4.978 (6)	-93.519	-4.646 (7)	-0.310	8.227 (7)
43	NS ₁ N ₂ -10	4.929	-87.626	-4.459 (9)	-0.299 (2)	8.157 (8)
44	NS ₁ N ₂ -11	4.871	-95.705	-5.882	-0.289 (1)	7.833
45	NS ₁ N ₂ -12	4.832	-70.885	-5.720	-0.357 (4)	6.798
46	NS ₁ N ₂ -5	5.075 (4)	-102.666	-6.108	-0.396	8.251 (6)
47	Control	3.458	-55.332 (2)	-3.907	-1.172	5.362

Table 40. Selection index values of mutant cultures of Nayagarh local on the basis of different selection criteria.

Sl. No.	Treatment Code	S.C - I	S.C - II	S.C - III	S.C - IV	S.C - V
1	NE ₁ -3	3.893	17.459	16.873	-0.203 (6)	6.382
2	NE ₁ -5	4.000	17.562	16.963	-0.160 (3)	6.518
3	NE ₁ -6	3.951	19.275 (8)	18.730 (9)	-0.181 (4)	6.665
4	NE ₁ -8	4.283	20.707 (2)	20.163 (2)	-0.221 (9)	7.149 (5)
5	NE ₁ -10	3.942	17.798	17.206	-0.145 (1)	6.463
6	NE ₁ -11	3.805	19.122 (9)	18.586	-0.455	6.493
7	NS ₁ -6	4.390 (5)	17.092	19.573 (5)	-0.303	6.755 (9)
8	NS ₁ -7	3.386	13.363	12.675	-0.433	5.386
9	NS ₁ -11	3.161	12.168	11.454	-0.269	5.100
10	NS ₂ -2	4.010	15.944	15.192	-0.194 (5)	6.329
11	NS ₂ -4	3.991	18.600	18.060	-0.370	6.403
12	NS ₂ -9	4.000	17.103	16.450	-0.345	6.421
13	NS ₂ -10	3.951	20.362 (4)	19.899 (4)	-0.312	6.811 (7)
14	NS ₂ -12	3.942	13.374	12.561	-0.313	5.910
15	NN ₂ -2	3.327	13.958	13.334	-0.412	5.429
16	NN ₂ -3	3.620	13.731	13.018	-0.443	5.661
17	NN ₂ -7	3.991	15.858	15.201	-0.415	6.249
18	NN ₂ -8	4.254 (7)	16.663	16.009	-0.332	6.575
19	NN ₂ -10	4.313	20.481 (3)	19.991 (3)	-0.304	7.126 (6)
20	NN ₂ -11	4.020	20.003 (6)	19.482 (6)	-0.298	6.810 (8)
21	NN ₂ -12	4.243 (9)	17.434	16.749	-0.351	6.722
22	NS ₂ N ₂ -3	3.805	19.484 (7)	18.973 (6)	-0.269	6.578
23	NS ₂ N ₂ -7	3.864	18.283	17.744	-0.151 (2)	6.457
24	NS ₂ N ₂ -9	3.912	18.258	17.683	-0.339	6.487
25	NE ₁ S ₁ -1	4.088	18.727	18.113	-0.274	6.721 (1)
26	NE ₁ S ₁ -2	4.625 (2)	20.165 (5)	19.579 (3)	-0.244	7.395
27	NE ₁ S ₁ -3	4.771 (1)	17.427	16.691	-0.214 (8)	7.165 (3)
28	NE ₁ S ₁ -4	4.030	16.027	15.286	-0.339	6.318
29	NE ₁ S ₁ -5	4.244 (8)	15.931	15.168	-0.247	6.508
30	NE ₁ S ₁ -6	3.825	15.197	14.496	-0.324	6.045
31	NE ₁ S ₁ -7	4.098	16.221	15.596	-0.211 (7)	6.398
32	NE ₁ S ₁ -8	4.556 (3)	18.891	18.256	-0.299	7.157 (4)
33	NE ₁ S ₁ -9	4.293 (6)	16.214	15.437	-0.377	6.561
34	NE ₁ S ₁ -10	3.932	17.124	16.821	-0.415	6.421
35	NE ₁ S ₁ -11	4.195	16.206	15.490	-0.435	6.791
36	NE ₁ S ₁ -12	4.293	20.918 (1)	20.306 (1)	-0.278	7.189 (2)
37	NS ₁ N ₂ -1	4.215	16.987	16.398	-0.377	6.578
38	NS ₁ N ₂ -3	3.703	16.332	15.627	-0.307	6.107
39	NS ₁ N ₂ -4	4.030	16.103	15.362	-0.429	6.324
40	NS ₁ N ₂ -7	3.571	13.838	13.113	-0.331	5.639
41	NS ₁ N ₂ -8	3.805	15.152	14.447	-0.303	6.034
42	NS ₁ N ₂ -9	3.844	13.147	12.340	-0.257	5.791
43	NS ₁ N ₂ -10	3.786	15.795	15.138	-0.333	6.100
44	NS ₁ N ₂ -11	4.137	16.444	15.703	-0.229	6.465
45	NS ₁ N ₂ -12	4.147	17.472	16.801	-0.362	6.525
46	NS ₁ N ₂ -5	4.391 (4)	14.882	14.073	-0.312	6.505
47	Control	3.044	11.033	11.447	-0.365	4.882

Table 41. Mutant cultures selected on the basis of different selection criteria with ranking of index values for each SC.

Treatment of code	Rank of selection				
	SC - I	SC - II	SC - III	SC - IV	SC - V
<u>Dhauri</u>					
DE ₁ S ₁ - 5	1	8	5	--	--
DE ₁ S ₁ - 6	3	9	--	--	3
DE ₁ S ₁ - 7	2	7	--	--	--
DE ₁ S ₁ - 11	9	--	--	9	--
DE ₁ S ₁ - 12	8	--	--	--	--
DS ₁ N ₂ - 7	5	--	8	3	--
DS ₁ N ₂ - 8	6	--	7	--	7
DS ₁ N ₂ - 9	--	--	9	2	8
DS ₁ N ₂ - 12	4	--	--	--	6
<u>Nayagarh local</u>					
NE ₁ - 8	--	2	2	9	5
NS ₁ - 6	5	--	5	--	9
NN ₂ - 10	--	3	3	--	6
NN ₂ - 11	--	6	7	--	8
NE ₁ S ₁ - 2	2	5	3	--	--
NE ₁ S ₁ - 3	1	--	--	8	3
NE ₁ S ₁ - 8	3	--	--	--	4
NE ₁ S ₁ - 12	--	1	1	--	2
NS ₁ N ₂ - 7	4	--	--	--	--

42. The expected genetic gain for the direct selection on yield (SC - I) was 0.6007, which was 13.08 percent of mean whereas, the same was 1.1068 for SC - II. The efficiency of SC - II was 84.32% higher over the direct selection. The efficiency of selection based on all the 8 characters was 13.69 higher than the direct selection and the expected genetic gain was 14.87% of the control. Similarly, the efficiency of selection of the mutant cultures of Nayagarh local was the highest (188.18%) in SC - III, where the yield components and yield were taken into consideration; followed by SC - II (180.35%) and SC - V (119.05%). The efficiency was 100% for direct selection. It was observed that the efficiency was highest in SC - II for Dhauri and in SC - III for Nayagarh local. The efficiency in both the sets of experiments was least for SC - IV, where, only morphological attributes were considered.

The average yield of the cultures, selected on the basis of five different selection criteria (SC) are presented in Tables 43 and 44. The average yield of Dhauri selected on the basis of yield per se (SC - I) was 10.73 q/ha as against 8.27 q/ha in the parent, there by indicating an increase of 29.74% over the parent. Similarly, the average yield of mutant cultures on the basis of SC - II, SC - III, SC - IV and SC - V were 10.82, 11.05, 10.61 and 11.63 q/ha raising the increase in yield to the tune of 30.83, 33.61, 28.29 and 40.62% of the parent respectively. The efficiency of SC - II, SC - III and SC - V were 3.66, 13.01 and 36.58% higher over the direct selection respectively.

Table 42. Expected genetic gain in yield and efficiency of selection on the basis of different selection criteria in M_4 generation.

	Selection criteria				
	S.C - I	S.C - II	S.C - III	S.C - IV	S.C - V
<u>Dhauri</u>					
1. Expected genetic gain in yield (g/plant)	0.6007	1.1068	0.6191	0.4314	0.6824
2. Expected genetic gain as % of mean (4.59 g/plant)	13.08	24.11	13.48	9.40	14.87
3. Efficiency over SC - I (%)	100.00	184.32	103.05	71.87	113.69
<u>Nayagarh local</u>					
1. Expected genetic gain in yield (g/plant)	0.4811	0.8676	0.9052	0.1209	0.5726
2. Expected genetic gain as % of mean (4.09 g/plant)	11.76	21.21	22.13	2.96	14.00
3. Efficiency over SC - I (%)	100.00	180.35	188.18	25.17	119.05

Table 43. Yield of mutant cultures of Dhauri selection on the basis of different selected criteria.

Sl. No.	Entry No.	Treatment code	Yield (Q/ha) of selected cultures based on selection indices				
			SC - I	SC - II	SC - III	SC - IV	SC - V
1	22	DE ₁ S ₁ - 5	11.40	11.40	11.40	--	--
2	23	DE ₁ S ₁ - 6	11.75	11.75	--	--	11.75
3	24	DE ₁ S ₁ - 7	9.32	9.32	--	--	--
4	27	DE ₁ S ₁ - 11	10.65	--	--	10.65	--
5	28	DE ₁ S ₁ - 12	11.52	--	--	--	--
6	41	DS ₁ N ₂ - 7	9.30	--	9.30	9.30	--
7	42	DS ₁ N ₂ - 8	11.65	--	11.65	--	11.65
8	43	DS ₁ N ₂ - 9	--	--	11.88	11.88	11.88
9	46	DS ₁ N ₂ - 12	11.25	--	--	--	11.25
10	Dhauri		8.27	--	--	--	--
Average yield (Q/ha)			10.73	10.82	11.05	10.61	11.63
Increase over parent (Q/ha)			2.46	2.55	2.38	2.34	3.36
Increase over parent (%)			29.74	30.83	33.61	28.29	40.62
Efficiency over direct selection			100.00	103.66	113.01	95.12	136.58

Table 44. Yield of mutant cultures of Nayagarh local selection on the basis of different selected criteria.

Sl. No.	Entry No.	Treatment code	Yield (Q/ha) of selected cultures based on selection indices				
			SC - I	SC - II	SC - III	SC - IV	SC - V
1	4	NE ₁ - 8	--	10.51	10.51	10.51	10.51
2	7	NS ₁ - 6	9.05	-	9.05	--	9.05
3	19	NN ₂ - 10	--	10.70	10.70	--	10.70
4	20	NN ₂ - 11	--	11.20	11.20	--	11.20
5	26	NE ₁ S ₁ - 2	10.60	10.60	10.60	--	--
6	27	NE ₁ S ₁ - 3	9.55	-	-	9.55	9.55
7	32	NE ₁ S ₁ - 8	10.70	-	-	-	10.70
8	36	NE ₁ S ₁ - 12	-	10.95	10.95	-	10.95
9	46	NS ₁ N ₂ - 7	10.72	-	-	-	10.72
Yield of Parent (Q/ha)			7.38				
Mean of the selected mutants			10.12	10.79	10.50	10.03	10.42
Increase over parent (Q/ha)			2.74	3.41	3.12	2.65	3.04
Increase over parent (%)			37.12	46.20	42.27	35.90	41.19
Efficiency over direct selection			100.00	126.46	113.87	96.71	110.96

Table 45. Performances of yield and yield attributes of different selected cultures in M₅ generation Var - Dhuli.

Entry No.	Treatment code	Plant height (m)	Maturity (days)	Pods/ plant	Seeds/ pod	100-seed weight (g)	Yield (Q/ha)
22	DE ₁ S ₁ - 5	60.57**	54.7*	29.40**	4.03	4.02**	11.40**
23	DE ₁ S ₁ - 6	51.23**	58.2*	26.20**	4.20	3.14	11.75**
24	DE ₁ S ₁ - 7	52.89**	52.7*	22.20**	4.90**	4.25**	9.33**
27	DE ₁ S ₁ - 11	38.90*	50.7*	24.80**	4.90**	3.70**	10.65**
28	DE ₁ S ₁ - 12	48.27	51.2*	28.00**	4.70**	3.60**	11.52**
41	DS ₁ N ₂ - 7	45.67	53.5*	29.00**	4.89**	3.50	9.30
42	DS ₁ N ₂ - 8	43.25*	51.7*	30.10**	4.20	3.70**	11.65**
43	DS ₁ N ₂ - 9	44.60*	53.4*	32.00**	4.00	3.43	11.88**
46	DS ₁ N ₂ - 12	45.23*	56.2*	30.00**	4.08	4.00	11.25**
Parent mean (control)		46.03	60.23	21.32	4.24	3.51	8.27
Mean of selected mutants		47.82	53.59	27.97	4.43	3.70	10.96
C.D. (0.05)		0.78	0.88	2.34	0.27	0.24	1.05
% of increase/decrease over parent		3.89	-11.02	31.39	4.48	5.41	32.52
Correlation with the index scores of							
	SC - I	--	--	- 0.2676	0.1283	0.3239	0.5570
	SC - II	--	--	- 0.8733**	0.3967	0.0177	0.0809
	SC - III	--	--	- 0.4764	0.2177	0.4077	- 0.1682
	SC - IV	--	--	- 0.7961**	0.4122	0.2276	0.4996
	SC - V	--	--	0.4402	- 0.4150	- 0.5835	0.2462

* Significant decrease at 0.01

** Significant increase at 0.01

Table 46. Performances of yield and yield attributes of different selected cultures in M₅ generation Var - Nayagarh local.

Entry No.	Treatment code	Plant height (m)	Duration (days)	Pods/plant	Seeds/pod	100-seed weight (g)	Yield (Q/ha)
4	NE ₁ - 8	43.5*	-63.20*	30.00**	4.11	3.30	10.51**
7	NS ₁ - 6	41.2*	-62.50*	28.00**	4.40	3.41	9.05**
19	NN ₂ - 10	46.1*	-60.70*	32.20**	4.12	3.20	10.70**
20	NN ₂ - 11	47.2	-60.80*	29.00**	4.20	3.10	11.20**
26	NE ₁ S ₁ - 2	48.1	-59.70*	28.00**	4.62	3.06	10.60**
27	NE ₁ S ₁ - 3	35.8*	-59.20*	24.50**	4.70	3.70**	9.55**
32	NE ₁ S ₁ - 8	41.3*	-56.80*	26.20**	4.50	3.30	10.70**
36	NE ₁ S ₁ - 12	47.2	-55.80*	30.00**	3.89	3.80**	10.95**
40	NS ₁ N ₂ - 7	42.5*	-60.30*	24.70**	4.76	3.42	10.72**
Parent mean		51.2	68.20	20.21	4.16	3.16	7.38
Mean		40.32	60.00	28.07	4.36	3.37	10.72
C.D. (0.05)		4.13	0.97	3.09	N.S	0.20	0.80
% of increase/decrease over parent		-21.25	-12.02	38.89	4.81	6.64	45.25
Correlation with the index scores of							
	SC - I	--	--	- 0.6043	0.6811*	0.2104	- 0.1820
	SC - II	--	--	0.8282**	0.7675*	- 0.2233	0.1262
	SC - III	--	--	0.8333**	- 0.7622*	- 0.1972	0.1560
	SC - IV	--	--	0.1118	- 0.0871	- 0.1816	0.1091
	SC - V	--	--	0.3150	-0.2324	- 0.0525	- 0.0992

N.S. Not Significant.

* Significant decrease at 0.01

** Significant increase at 0.01

The average yield of the cultures of Nayagarh local selected on the basis of yield per se was 10.12 q/ha as against 7.38 q/ha in parent and thereby indicating an increase of 37.12% over the parent. Similarly, the average yield of the cultures selected on the basis of SC - II, SC - III and SC - V were 10.79, 10.50 and 10.42 q/ha showing an increase of 46.20, 47.27 and 41.19 percent over the parent respectively. The relative efficiency of SC - II, SC - III, SC - IV and SC - V were 124.46, 113.87, 96.71 and 110.96 percentages respectively over the direct selection (SC - I).

The efficiency of the different selection criteria were also assessed on the basis of relationship between the index scores of the selected mutant cultures and it's performances during M_5 generation and presented in Tables 45 and 46. It was revealed that significant correlation was observed between the index scored and pods/plant for the selection criteria of Sc - II and Sc - IV in Dhauri, whereas, it was significant for SC - II and III for the cultures of Nayagarh local. Though the relationship between seeds/pod and index score was insignificant for all the selection criteria of the mutant cultures of Dhauri, the same trait was significant for SC - I, SC - II and SC - III for the cultures of Nayagarh local.

4.9 M_5 GENERATION

The performances of yield attributing characters of the selected mutants during M_5 generation of both the varieties are presented in Tables 45 and 46. The range of variation among mutant cultures of Dhauri for pods/plant, seeds/pod, 100-seed weight and plant yield varied from 22.20 - 30.10, 4.00 - 4.90,

3.14 - 4.25 and 9.33 - 11.88 respectively whereas, in the respective traits in the parent variety were 21.32, 4.24, 3.51 and 8.27. All the selected mutant cultures should^e show significant reduction in maturity period over parent. Three mutant cultures showed significant increase in plant height, while, four mutant cultures showed significant decrease in plant height.

The range of variation observed in the selected mutant cultures of Nayagarh local varied from 24.50 - 30.00 for pods/plant, 3.89 - 4.70 for seeds/pod, 3.10 - 3.80 for 100-seed weight and 9.05 - 11.20 in yield/ha., while, these characters in the parent variety were 20.21 4.16 3.06 and 7.38 respectively. All the selected mutant cultures of Nayagarh local showed significant reduction in duration, while seven out of nine selected mutants observed significant reduction in plant height.

All the selected mutant cultures selected from Dhauri were developed from combined mutagenic treatments and the occurrence of EMS + SA combination was most frequent. In Nayagarh local, five out of nine were selected from combined mutagenic treatment. The EMS + SA combination was found best over rest of the treatments in producing superior mutant cultures.

CHAPTER - V
DISCUSSION

DISCUSSION

Greengram, a major pulse crop of Indian sub-continent, ranks third to chickpea and pigeonpea. It contributes 15% of the total pulse production. Sometimes, reservations are expressed that there is a limited range of genetic variability in mungbean (Ahmed et al., 1978). Further, non-availability of doners for some of the productive characters pose another problem in combining such characters for better yield. This view had led the breeders to go for mutation breeding to induce polygenic variation and to select the desired micro-mutants. Sigurbjornson and Micke (1969) were of the view that mutation method could be useful with regards to direct release of mutants and use of induced mutants in recombination breeding.

The usefulness of micromutations was established by Gregory (1956, 1961). The development of about one thousands varieties in 98 species enhanced the significance of induced mutation research in plant improvement (Micke, 1990). In spite of a great success in the use of induced mutation, the breeders still depend a large extend on the chance occurance of mutation. But then, there is scope of getting better mutant lines through adoption of suitable methodology and following appropriate screening techniques (Khan, 1984). In the present investigation the mutation programme was taken up as a tool to induce variation in two varieties of greengram and to develop suitable cultivar(s) particularly suitable for the rabi season.

In addition to use of single mutagenic treatment, physical chemical combine treatments were used successfully by several workers in this crop (Grover and Tejpaul, 1979); Krishnaswamy et al., 1977; Bahl and Gupta, 1982 and Khan, 1983). Reports on chemical combinations of mutagen were meagre (Chopra and Swaminathan, 1966; Kak and Kaul, 1975; Mohapatra et al., 1987). In the present investigation both single and combine treatments of three chemical mutagens were used and it was revealed that there was reduction in germination, survival, seedling height, root length, shoot length and pollen fertility in M_1 generation. These reductions were more drastic in combined treatment of chemical mutagens as compared to the single mutagenic treatment. This may be due to synergistic effect of the chemicals. Further, these parameters showed stimulatory effect at lower dose and inhibitory action at higher dose for all the traits except germination and survival. The stimulatory action might be due to breaking of unfavourable physiological conditions of seeds and the inhibitory action at some concentrations might be due to damage of DNA and meiotic disturbances. This condition was in accordance with the observation of Sahoo and Samolo, (1973); Chandra & Tewari, (1979); Mohapatra (et al. 1987); as well as Ignacimuthu, (et al., 1989).

The pollen and seed sterility were higher under higher concentration, confirming the dose dependency of fertility and agreed to the findings of Chang and Hsich, (1957); Yamaguchi (1963); Sharma (1970); Chakrabarty (1975); Khan and Hashim (1979). The reduced fertility at higher concentration might be

due to low frequency of gross structural change and other meiotic disturbance. Combined treatment of chemical mutagens caused more reduction in fertility than the single chemical treatments. This was in confirmity of the findings of Ramaswamy, (1973) in blackgram; Krishnaswamy, (1983) in greengram and Shrinivasan, (1977) in redgram. Sato and Gaul, (1967) while studying the effects of EMS induced sterility in barley reported that seed sterility occurred in a very low frequency of translocation due to cryptic chromosomal deletion and ruled out the genetic change (point mutation) as a cause of sterility.

The root growth was slower than shoot growth indicating root system to be relatively more sensitive to mutagens. This could possibly be due to inhibition of cell division and cell elongation in the root cells. The shoot growth was reported to be due to cell elongation while, root growth was more dependant upon cell division. Ivanora, (1968); Mishra, et al., (1980); Sinha and Godward, (1968) also observed delay in rooting than shooting following irradiation.

The range of variation of the observed characters of the treated populations of both the varieties was more enlarged from the respective controls which might be due to application of mutagenic treatments and was in agreement with the findings of Dahiya, (1973); Prasad, (1976); Mishra et al., (1980); Singh et al. (1980); Mohapatra et al., (1983); Kadapa, (1985) and Khan, (1988). The nature and amount of variability induced after chemical treatment in different quantitative traits in M₁ generation indicated that means of most of the characters in

most of the treated populations of both the varieties were significantly decreased whereas, in some of the treated populations it was significantly increased from the control. The reason for reduction in mean might be due to deleterious effect of mutagens (Mujeeb, 1974; Rajput and Khan, 1971; Swaminathan, 1973; and Khan, 1984, 1988). All the quantitative characters in M_1 were more adversely affected in combine treatments. This might be due to synergistic effect of mutagens towards negative side for some of the characters which corroborated the findings of Reddy et al. (1973) in rice.

Relationship between M_1 parameters with M_2 mutation frequency

Some of the research workers expressed their views that the chromosomal or gene mutations induced in M_1 by mutagenic treatment are likely to be transferred to the next generation (Bender & Gaul., 1966, 1656). But Gaul, (1977) reported that physical damage caused by the mutagenic treatment remains in the M_1 generation only. Bender and Gaul (1965, 1967) had observed that a part of the induced sterility was due to gene mutation or due to chromosomal or cytoplasmic mutation or due to physiological change. The induced gene, chromosomal a cytoplasmic mutation might be transferred from M_1 generation to the next generation, while physiological damages were retained in M_1 generation only. Therefore, it was felt necessary to find out the relationship between M_1 damage and M_2 mutation frequency to identify effective mutagenic treatment. The present study indicated that M_1 parameters like germination, survival, seedling height, pollen and seed sterility had a significant and positive correlation with chlorophyll mutation

frequency in both the sets of experiments. Ehrenberg, et al., (1961); Sahoo and Samolo, (1973) as well as Mishra et al., (1981) observed that M_1 pollen sterility and pollen size variability had significant positive correlation with M_2 mutation frequency in greengram and, therefore, could safely be taken as criterion to identify effective mutants. Sharma and Bansal (1970) observed highest rate in medium fertility group in M_1 plants, while Kunzel et al., (1971) noticed linear increase in M_2 mutation frequency with increase number of sterile plants in M_1 . Thus, the present finding was in conformity with findings of the above mentioned research workers. Therefore, M_1 pollen sterility, seed sterility, seedling height, survival and germination could be taken as index to find M_2 mutation frequency.

There are few earlier reports on the correlation between M_1 parameters and M_2 population variance to give stress on specific M_1 parameters to identify the micromutations. It was observed that seed sterility, survival percentage and pollen sterility percentage showed higher positive significant correlation with the population variances of some traits in both the varieties. Only seedling height in some cases showed negative significant correlations with population variance in both the varieties. Hence, identification of mutant population based on these M_1 parameters might yield fruitful results.

Macro- and Micro-mutation in M_2 generation

A broad spectrum of mutant type plants were observed in the treated populations of the present study in M_2 generation following mutagenic treatments. A high proportion of

chlorophyll mutants were reported by Dahiya, (1973); Grover and Tejpaul, (1979); Singh et al., (1979); Singh and Chaturvedi, (1980); Bahl and Gupta, (1982); Krishnaswamy, (1983); Grover and Virk, (1984); Rao et al., (1984) and Khan, (1985). The chlorophyll mutation frequency in M_2 is the most reliable measure for evaluation of mutagenically induced genetic alternation. In the present study, five different types of chlorophyll mutants were isolated in M_2 generation. Those were chlorina, xantha, albina, viridis, and sectorial. Dahiya, (1973); Grover and Tejpaul, (1979); Khan, (1981); Bahl and Gupta, (1982); Krishnaswamy, (1983); Rao et al., (1984) and Sahoo, (1994) have earlier reported similar type chlorophyll mutants in greengram. The occurrence of chlorina type was most frequent and sectorial type the least in both the varieties. The NG treated population showed more chlorophyll mutants followed by SA and EMS in descending order in both the varieties. More number of chlorophyll mutants were found in combine treatment than in the single mutagenic treatment. The specificity of different mutagens to produce different types of mutation was well established. It was revealed from the present investigation that different varieties of greengram differed in their capacity to produce different types of chlorophyll mutants, which was in conformity with the findings of Khan, (1981); Mishra and Samolo, (1982) and Krishnaswamy, (1983). The varietal differences to mutagen as described by Krishnaswamy, (1983) was due to level of differentiation of the apical and lateral meristem. Those that have already differentiated at the time of mutagen treatment were expected to respond differently

from those that were yet to be differentiated (Blixrt, 1972). Rosichan et al., (1983) have reported the species differences in azide sensitivity, which was subsequently supported by Rines (1985) who observed varietal difference was due to difference in uptake or metabolism of the mutagens, cellular repair mechanism or the site of specificity and mode of action of mutagen.

Mutations that alter morphological characters of greengram have been reported by Dahiya, (1973); Chaturvedi, (1975); Yadav and Singh, (1983); Grafia and Santosh (1979); Pande and Raghubansi, (1988). In the present study morphological mutants observed in M_2 generation were trifoliate, quadrifoliate, pentafoliate, early flowering, late flowering, tall plant, dwarf plant, profused branched plant, trailing, hairy podded plant etc. Foliar mutations observed in the present investigation were in agreement with the earlier findings of Bandopadhyay and Bose, (1983); Krishnaswamy, (1983); Khan et al., (1989) and Satyanarayanet al., (1989). It was noticed that NG treated populations yielded high morphological mutation frequency followed by SA and EMS in Dhauli, while it was highest in NG treated population followed by EMS and SA in Nayagarh local. This might be due to drastic action of NG, which was considered as super mutagen by Rapoport et al., (1966) and Swaminathan (1968).

The efficiency of mutagen in inducing morphological mutations were in order of SA > NG > EMS in Dhauli and NG > EMS > SA in case of Nayagarh local. Thus, higher frequencies of morphological mutations were induced by the treatment with NG

and SA might be due to their efficiency in inducing more of point mutation as reported by Bautz and Freese, (1960); Nilan *et al.*, (1973). The differential response of mutagens observed in the present findings might be due to the fact reported by Rines (1985). Similar differential response of mutagenic treatments have also been reported by Swaminathan *et al.*, (1968), Rao and Jana, (1976 a and b), Tickoo and Jain, (1980), Shaikh *et al.*, (1983), Tickoo, (1987), Shaikh (1990) in different crops. The total mutations in different mutagenic treatments revealed that there was an increase in mutation frequency with increase in dose. Such dose dependant findings agreed to the earlier reports of Sahoo and Samolo, (1973); Sharma *et al.*, (1979); Mohapatra *et al.*, (1983) in greengram. The response of the two varieties to different mutagens in producing macromutation was different. This indicated that frequency of mutation was governed by the genetic architecture of the material used and confirms to the earlier findings of Chao and Chai, (1961); Davis, (1962); Mishra and Samolo, (1982); Krishnaswamy, (1983); Reddy and Prabhakar, (1983); Rosichan *et al.*, (1983); Iqbal and Khan, (1989) and Sahoo, (1994) in different crops.

Mean and variability of the quantitative characters in M₂ generation

Mutations affecting quantitative characters can be best assessed by estimating mean and genetic parameters in the mutagen treated population. It has been experimentally proved that induced mutation could increase yield as well as other quantitative characters in crop plants (Khan, 1981, 82, 83, 84;

Verma and Singh, 1984). The effect of the micromutation are small and often confounded with environmental variation. Their presence could not be recognised from a single plant but could be detected in a group of plants either by eye estimation or by measurements (Gaul, 1964). Mutation that caused variation in quantitatively inherited characters have been identified in greengram by Dahiya, (1973); Singh and Chaturvedi, (1981); Kundu and Singh, (1982); Khan, (1982, 1983, 1984, 1985); as well as Verma and Singh, (1984). Induced micromutations are generally low and associated with less defective changes and hence more readily exploited in breeding programme.

In the present investigation change in the mean values of the treated populations of both the varieties in all phenotypically observed characters occurred both in positive and negative directions, most of them were significantly different from control. The trend was positive for majority of the treated populations with regards to pod length, whereas it had both positive and negative shift for rest of the traits in case of the variety Dhauri. The later condition mostly prevailed in the variety Nayagarh local for all the characters. Dahiya, (1973); Rajput, (1974); Khan, (1981); Seth et al., (1984); and Khan, (1985) have reported a negative shift in the mean, whereas a positive shift of mean in greengram was reported by Rajput, (1974); Prasad, (1976); Khan, (1981, 1982, 1984, 1985, 1983); Seth et al., (1983, 1984); Singh et al., (1988). The reason for decrease in mean of the treated population as compared to control could be due to high sterility influenced by the direct effects and inductions of

more mutations with negative effects (Khan, 1982). The shift in mean in a positive direction could be due to occurrence of plus effect (Rajput, 1974; Khan, 1981). Further, it was observed that there was no definite trend in the change of mean in relation to dose or mutagens. This might be due to creation of critical treatment level on which more number of mutations were produced than other treatments. Oka et al., (1958) reported that want of a clear cut change in mean in same trait could be due to mutations of major and minor genes in both directions. Further, it was observed that change in mean of the M_2 treated population differed with mutagen used, their concentration, character chosen and variety used which was in confirmity with the observations reported by Khan (1981).

Induced variability in the treated populations for polygenically controlled characters was reported by Dahiya, (1973), Kundu (1980), Singh et al. (1980), Kundu and Singh, (1982) and Khan (1984, 1985). In the present investigation, significant increase in variation over control was observed in the treated populations for all the characters in both the experiments indicating scope for selection. The magnitude of variation of the treated populations also varied with mutagens used, characters chosen and parental material. The increase in variability in this study could be explained to be due to mutations of polygenes governing quantitative characters which was in confirmity with the findings of Khan (1988). In the present study increase in variance did not result corresponding increase in mean which was in agreement with the findings of Oka et al. (1958). These characters were found to respond

differently to mutagen. No linear relationship was observed between dose and variance. This could be due to additive uncontrolled environmental variation.

Efficiency of single and combined treatments

It has been the practice to draw conclusion on mutagenic efficiency from the data on viable mutants, which probable is not always correct (Rao and Natrajan, 1965; Goud, 1968). Hence, mutagenic efficiency was calculated by taking into consideration the chlorophyll mutations, because such mutants could easily be classified and scored with much accuracy (Gaul, 1977; Nerker, 1977; and Khan and Hashim, 1979). Mutagenic efficiency was higher at lower concentration of mutagen and agreed to the findings of Nerker (1977), Prasad (1972) and Khan and Hashim (1979). The greater efficiency at lower concentration of mutagens was due to the fact that the biological damage increases with increase in dose Konzak (1965). Rao and Natrajan (1965) reported that mutation to be realised, the mutated gene must be able to reproduce itself. On this basis SA appeared to be more efficient mutagen for Dhauli and EMS was for Nayagarh local, in single mutagenic treatment. The combined treatment of chemicals were found to be more efficient than single treatment. This might be due to the possible synergistic action of two mutagens used. From the practical point of view higher frequency of mutants obtained has been adopted as the best measure for comparison of mutagenic efficiency (Kawai, 1969). Similar views were expressed by Ehernberg (1960), Rao and Natrajan (1965).

The problem in micromutation breeding lies with the

quantum of sustained work over generations to identify superior high yielding mutants from a large number of similar looking plants Sharma (1986) and Mohapatra (1987). Therefore, it is necessary to select micromutants from the early generation to reduce the work load.

Most of the studies on induce polygenic variation showed differences with respect to generation from which selection process should start. After the studies of Brock (1965, 1967) it became common practice to advance the normal looking M_2 plants to M_3 generation and apply first dose of selection in M_3 . This resulted increase volume of nonmutated plants and delay in isolating promising variants. Thus, the proclaimed advantage of rapid progress in breeding through induction as compared to hybridization is lost. Selection in M_3 generation was reported by Papa et al. (1981), Miah et al. (1965), Bhatia and Van doer Veen (1965), Gaul et al. (1969), Jana and Roy (1973), Ravi and Singh (1979) Mohapatra (1983) and Sahoo (1994) in different crops. Comparison studies of selection in M_2 and M_3 generation made by Scossiroli (1968) and Sharma (1977) indicated that for certain traits selection in M_3 was more effective whereas, for others selection in M_2 was effective. Oka et al. (1958) suggested the use of bulk method upto M_4 generation and selection in M_5 . Tickoo and Jain (1979), Bhadra (1982), Kharkwal (1983) concluded that promising progenies can be identified with high degree of confidence in M_2 on the basis of mean and variance. Selection in M_2 is more effective than M_3 (Palézonⁿ, 1966; Jana & Roy, 1973). This was most probably because of materials selected once in M_2 was confirmed with

higher probability in subsequent generation (Ravi et al. 1979). Even if the materials selected in M_3 or later generations has a higher probability of getting fixed as promising strain. Selection in M_2 and subsequent selection in M_3 and M_4 generations were reported by Gregory (1955), Borojeric (1965), Sharma (1977), Tickoo and Jain (1980), Sharma (1983), Sinha & Bharati (1990), Mishra (1992) and Sahoo (1994) on different crops and reported useful selection of micromutants.

Thus, irrespective of characters shows increase in variance with the advancing generation or not assessment of variability in early generation should be a great help in reducing the volume of work and save time. Sharma (1986), reported that selection of variants in M_2 will bring micromutations almost at par with macromutations.

Therefore, attempts were made to explore the possibility of selecting for polygenic variability in M_2 .

Genetic variability

Genetic variability in the expression of a quantitative character is dependant upon a range of genetically controlled variable traits under consideration. Therefore, an attempt was made to assess the nature of variability induced due to mutation in the selected cultures during M_4 generation. In general, phenotypic coefficient of variability (PCV) and genotypic coefficient of variability (GCV) were moderate for all the traits except days to maturity and yield/plant of the cultures of Dhauli, whereas low for all the traits except plant height and pods/plant of Nayagarh local. It indicated that the variability was induced for all the traits to some extent by

mutation. This finding, in general, agreed to the findings of Kundu and Singh (1982); Rao (1982); Khan (1982, 1984, 1985, 1988); Seth and Choudhury (1983) and Verma and Singh (1984) who affirmed the presence of genetic variability in the size and structure of quantitatively inherited characters after induction of mutation in greengram. Low variability induced amongst the cultures of Nayagarh local might be due to less sensitivity of the local variety to the mutagens.

Heritability provides a quantified estimate indicating the relative importance of genetic and environmental components in the expression of that trait. In the present investigation high estimates of heritability was observed for all the traits in both the sets of experiment indicating major part of the phenotypic variability induced through mutation might be due to genetic cause. This finding was in agreement with the findings of Choudhury et al. (1980); Mishra et al. (1982); Ananda Kumar et al. (1986); Samala (1986) and Khan (1988). High heritability estimates coupled with high genetic advance was more viable than heritability alone in predicting the effect of selection (Johnson et al., 1955). The estimation of genetic gain expressed as percentage of mean was high for pods/plant and medium for plant height, seeds/pod, days to maturity and yield/plant in the cultures of both the varieties. This confirmed to the findings of Bhargava et al. (1966), Veerswamy, et al., (1973), Parida and Singh, (1985) and Khan (1988) in greengram. In the present study, estimation of high heritability along with high genetic advance was observed for pods/plant in the cultures of Dhauri. High heritability along

with medium genetic advance was observed for pods/plant in Nayagarh local indicating better response for selection of this trait (Rangaswamy et al., 1984). Heritability estimates together with genetic advance indicates the mode of gene action for the character (Panse, 1957). The association of these two estimates indicated additive type of gene action for pods/plant, whereas nonadditive type of gene action for rest of the traits.

The correlation coefficient provided a measure of relationship between traits and serve to assess the chance for mutual improvement of two characters by a common selection. Though, it was reported by many workers that there exists associations between yield and the component traits, it was reported by Sakai and Suzuki (1964) that induction of mutation helped on weakening or strengthening or altering character association. In addition to this, correlation between characters depends upon joint and individual action of selection, linkage and pleiotropy. Therefore, an attempt was made to study the character association between yield and it's component traits in two sets of the mutant cultures and it was observed that correlation coefficient between yield was significant with number of pods/plant, branches/plant and yield/plant in Dhauli. It was significant between yield and pods/plant in Nayagarh local indicating selection based on such traits could improve yield. The association at phenotypic level was lower in most of the pairs of characters indicating strong inherent association between them. A non-significant association among some pairs of characters indicated that the

scope of improvement could be achieved by independent selection. These findings, in broad sense, were in agreement with the findings obtained by Sandhu et al., (1978); Mishra et al. (1982) and Kumar and Reddy (1987). Genotypic and phenotypic correlation coefficients for some pairs of characters showed wide difference. In Dhauli, the correlation coefficient was wider between days to flowering and days to maturity days to maturity and branches/plant; days to flowering and branches/plant; branches/plant and 100-seed weight; 100-seed weight and pods/plant. In Nayagarh local, this difference was wider between days to maturity and branches/plant. These results corroborated to the earlier findings of Rao and Siddiq (1976), Sharma and Sharma (1981), Mohapatra and Singh (1988). These finding indicated that mutagenic treatments were found to be effective in changing the character association patterns at least in some of the traits.

In a selection programme, to increase seed yield based upon correlation studies, it should be considered that selection to increase one trait does not lead to deterioration in another trait. It was observed in the present investigation that in both sets of the experiments the direct contribution of pods/plant to the seed yield was highest. It was followed by seeds/pod and 100-seed weight in Dhauli and 100-seed weight and seeds/pod in Nayagarh local. The data confirmed the observations from correlation analyses that large number of pods/plant was a major contributing factor for high seed yield. As the direct effect was too high for pods/plant the indirect effects via other characters tend to be small or often

negative. Direct effect to yield/plant through seeds/pod was high and positive. But the positive contribution was smaller than pods/plant. The indirect contributions of seeds/pod via other characters were both positive and negative. The direct contribution of seed weight to yield/plant was also positive and high. The potential benefit of seed yield by selection for large seeds was nullified by large negative indirect effect via pods/plant.

In the present investigation, negative direct effect on yield were reported for days to flowering and plant height in both the experiments. Days to maturity and branches/plant showed positive and negative direct effect in both the varieties. The reasons for both positive and negative contributions for the above said characters could not be explained without additional information on genetic material and environment (Satyan et al., 1982). The findings obtained, in general, agreed to the findings of Sandhu et al. (1973), Malik et al. (1985); Khan (1985); Giriraj and Vijoy Kumar (1974); Boomikumaran and Radhinam (1981); Singh et al. (1978). Over all, the path coefficient analyses supported giving major selection efforts to pods/plant and seed size.

Genetic divergence

Understanding the magnitude of genetic divergence for yield components is essential in planning the breeding programme. It helps in choosing parent combinations to get maximum recombinations and also in understanding the pattern of evolution for different characters. The extent of genetic divergence to yield and it's related morphological characters

in mungbean have been investigated and evaluated by many research workers (Gupta and Singh, 1970; Malhotra et al. 1974; Ramanujam et al. 1974; Singh et al. 1976; Ghaderi et al. 1979; Malik et al. 1985; Ramana and Singh, 1987; Bhadra and Akhtar, 1991. Use of multivariate analysis to classify the different micromutations into different groups have been reported by Mohapatra et al. (1983), Baisakh (1985), Mohapatra (1987) and Sahoo (1994) in greengram. This multivariate analysis takes into consideration several quantitative traits simultaneously for which it is considered as a dependable tool to determine suitable difference among micromutations. The present study aims at quantification of the magnitude of genetic diversity for yield and its component traits as well as some morphological characters of different mutant cultures, originated from different chemical mutagenic treatments.

Analysis of variance revealed that the difference among the mutant cultures in both the sets of experiments were highly significant for all the characters under study indicating presence of notable genetic variability between them. Further test based on Wilk's criterion showed that the difference among the cultivars in respect of pooled effect of all the eight characters were significant.

Grouping of mutant cultures

Forty seven mutant cultures isolated each from two varieties of greengram were grouped into 11 and 12 clusters for Dhauli and Nayagarh local respectively. This indicated diversified type of genetic variability induced by the mutagenic treatments. It was revealed from the clustering pattern that

grouping of clusters usually did not follow in accordance with the source of origin. In the present investigation, Nayagarh local alone formed a separate cluster (cluster VI) in one set, whereas in another set Dhauli along with other two mutant cultures induced by different chemical mutagens formed another cluster. Mutant cultures isolated from different mutagenic treatments were also grouped under a single cluster in both the sets of experiment. On the contrary, in most of the cases, cultures developed from same mutagenic treatment were grouped into different clusters and no regular pattern was observed. This finding agreed to the findings of Mohapatra et al. (1983, 1987); Baisakh (1985); Samal (1986) and Sahoo (1994) in greengram. The cultures developed from combined treatments of mutagens were grouped into different clusters indicating the combine mutagenic treatments were much effective in inducing micromutations of greater divergence. The cultures which evolved from a common habitat also had ample genetic diversity and fell into different clusters. The substantial variability among the cultures, which evolved from the same treatment could be either due to existence of heterogeneity by marginal out crossing or through balance polymorphic system or directional selection pressure applied to achieve the goal as suggested by Katiyah and Singh (1990), Murty and Arunachalam (1966). Upadhaya and Murty (1970) pointed out that genetic drift or selection can cause high diversity among the races than the geographic isolation alone. Das et al. (1973), however, thought that if similar selection criterion are exercised, it may result in genetic similarity of the cultures developed by

different mutagenic treatments. Hence, the present findings were in agreement to the above findings.

An assessment of contributions of different characters revealed that branches/plant, days to maturity, plant height and 100-seed weight contributed maximum to the total divergence in descending order in Dhauri. Of the eight characters, only the first three contributed about 48% to the total divergence indicating major role in maintaining variability in the treated populations. In case of Nayagarh local, 100-seed weight contributed maximum to the total divergence followed by days to maturity, branches/plant and days to flowering indicating their major role in creating variability. The primary factors contributing to genetic divergence have been reported in various pulses including mungbean. Gupta and Singh (1970), reported that seed index and pod length had maximum contribution to total divergence, which is in contrary to the present findings might be due to different types of genotypes. However, narrow range of variability was observed for number of branches/plant, number of seeds/pod, pod length and 100-seed weight by them. Malhotra et al. (1974) and Ramanujam et al. (1974) reported maximum contribution of days to flowering, seed size and primary branches towards genetic diversity in greengram. The present finding in general, agreed to the findings of the above mentioned research workers. Differential selection for plant structure branches/plant, 100-seed weight and maturity might have resulted a greater variation for those traits, which ultimately contributed maximum to the total diversity.

In the mutant cultures of Dhauli, based on the cluster mean, it was revealed that highest yield was observed on the cultures of Cluster XI. The cultures of this cluster were short in height, medium duration types associated with high number of pods/plant, seeds/pod and 100-seed weight. The parental type falls in cluster II along with other two cultures and possessed low yield compared to all other clusters. Next to cluster XI other clusters having high yield were cluster IX and III in descending order. Cluster mean indicated that selected mutants, besides having short duration type also had high yield and superior yield attributes. Therefore, the two cultures i.e. DE-3 from cluster IV and DE₂N₂-2 from cluster V, as they having high inter cluster distance besides desirable characters, may be selected for hybridization programmes, so that better recombinants with desirable genes can be obtained. Similarly in case of Nayagarh local the parental type fell in cluster VI alone and had the lowest yield among all other clusters. Cluster XI had the highest yield followed by cluster XII and VIII. Culture NS₁-6 from cluster II and NE₁S₁-12 from cluster VII as they were having high inter cluster distance besides short height and high yielding ability may be selected for further breeding programme, so that transgressive segregants can be obtained.

Selection indices

The success of mutation breeding for improvement of polygenic traits depends not only on the quality of induced mutation but also on the ability of the breeder to select useful micromutants. In order to increase the chance of success in selection, the use of multiple criteria selection in later

generation has been advocated by Smith (1936), Lush (1940), Banarjee et al. (1976), Das (1978), Zakri et al. (1983), Devkota (1987), Mishra (1992) and Sahoo (1994). This technique of discriminant function offers an effective method to construct selection indices. Palenzona (1966) and Khan (1984), compared the efficiency of selection for M_2 and M_3 generation, whereas Oka et al. (1958) suggested the use of bulk method upto M_4 generation and selection in M_5 . Mishra (1992) indicated that selection in M_3 or latter generation would increase the volume of material to be handle of which a major portion might be non-mutated one and may delay in process of isolation of promising varieties.

In the present study a systematic approach was undertaken for selecting micromutants with higher yield. In the process emphasis was given on rigorous rejection of nonmutant types in M_2 and M_3 generation. In M_4 generation mutant cultures were identified on the basis of yield per se. and were compared with index scores of different selection criteria. It was revealed that all the multiple criteria selection were superior over the direct selection excepting SC - IV as expected genetic gain were higher for those culture for both the sets of experiment. Multiple criteria selection on the basis of SC - II was highest for the cultures of Dhauri whereas, of SC - III for the cultures of Nayagarh local. The efficiency of SC - IV could not prove superior over the direct selection in both the varieties, might be due to the inclusion of characters in it which were negatively correlated with yield. On the other hand in both the sets of experiment the rest of the selection criteria were

found superior over direct selection. Earlier studies indicated that selection would be effective with multiple criteria selection on pods/plant, seeds/pod and 100-seed weight (Mishra, 1992 and Sahoo, 1994). Thus, the expected efficiency of different selection criteria observed in the present study were in confirmity with the findings of the above mentioned research workers.

The realised yield improvement in M_5 generation indicated that multiple criteria selection on the basis of SC - II, SC - III and SC - V were more effective than the selection on the basis of yield per se. (SC - I). Finally, the mutant cultures which were developed from by combined treatments in both the varieties excepting four of the Nayagarh local were found productive and possessed superior yield attributing characters. Therefore, the selection procedure applied in this study was effective in identifying superior micromutants. In general, it was observed that EMS + SA combined mutagenic treatment was effective in producing superior mutants with high yield.

CHAPTER - VI

SUMMARY AND CONCLUSION

SUMMARY & CONCLUSION

The present investigation on "Studies on independent and combined mutagenic treatments on induction of mutation in greengram" was taken up to induce polygenic variability in quantitative characters in greengram and to selected desirable mutants for yield improvement of this crop. Two adopted varieties namely Dhauli and Nayagarh local were selected for this study. The seeds of these varieties were administered with six different concentrations of single and twelve combined mutagenic treatment of three chemical mutagens, i.e., Ethyl methane sulphonate (EMS), Sodium azide (SA) and Nitroso guanidine (NG). The single mutagenic concentration were 0.4 and 0.2% of EMS (E_1 , E_2), 200 ppm and 100 ppm of SA (S_1 , S_2), 200 and 100 ppm of NG (N_1 , N_2). The different combined treatments of mutagens were E_1S_1 , E_1N_1 , S_1N_1 , E_2S_2 , E_2N_2 , S_2N_2 , E_1S_2 , S_1N_2 , E_2S_1 , E_2N_1 and S_2N_1 .

In case of single mutagenic treatments, seeds presoaked for 12 hours were treated for 6 hours whereas, the treatment duration was 3 hours in each mutagen for combined treatments. The control seeds were also presoaked for 12 hours in distilled water before sowing. Two separate trials were conducted in RBD for two varieties during M_1 generation. Seeds from two fruiting bunches from all M_1 plants were bulked treatment wise and grown

in M_2 generation. Twelve plants (20% of 60 M_2 plants) were selected from each treatment on the basis of higher yield and the progenies of these selected plants were grown in M_3 under compact family block design. The yield performance between treatments and between progenies within treatment were studied. Forty six mutant cultures were selected from each of the sets on the basis of yield. These selected mutants along with the respective control were evaluated in M_4 generation. The variation, correlation among yield attributes, the direct and indirect contribution of characters to yield, their divergence and scope for selection through different multiple selection criteria were estimated during this generation. The selected mutant cultures were again evaluated during M_5 generation to assess, the efficiency of five different selection criteria (SC).

M_1 GENERATION

All the mutagenic treated populations recorded reduction in germination, survival, root and shoot length, seedling height, seed and pollen sterility from the control in both the varieties. These reductions were more spectacular in combine mutagenic treatments than single treatments. Root length was more affected than shoot length. In general a dose dependent reduction in the above characters was observed. In majority of the cases, these reductions were more pronounced in NG treated populations followed by SA and EMS.

Both increase and decrease in mean of the eight quantitative characters were recorded in the M_1 treated populations from control. The variability was more in all the

treated populations. Treated populations of both the varieties showed less recovery index (RI) than control. RI of the combine treatment of mutagens was lower than single mutagenic treatment. The coefficient of interaction was additive in all the combine treatments of both the varieties, except E_2S_2 , S_2N_2 , E_1S_1 , E_1N_1 , S_2N_1 and S_1N_2 treatments of Nayagarh local.

M₂ GENERATION

Five different chlorophyll mutations like Xantha, albina, chlorina, viridis and sectorial were observed in both the varieties. In the treated populations of ~~treated with~~ NG showed more chlorophyll mutation followed by SA and EMS in Dhauli and EMS and SA in Nayagarh local. Different morphological mutants observed were tall, dwarf, early, late, quadrifoliate, pentafoliate, profuse branching, trailing, hairy podded plant spreading type etc. The efficiency of mutagens in producing different types of morphological mutations were in order of SA > EMS > NG in Dhauli and NG > EMS > SA in Nayagarh local. The frequency and spectrum of mutation varied with mutagens, their concentrations, methods of application and variety.

The mutagenic treated populations showed shift in mean in both directions but increase in variance as compared to control for eight quantitative characters studied. The change in mean and variance varied with mutagens, their concentrations, characters chosen and variety used. Increase in variance did not result in corresponding increase in mean for most of the traits studied.

M₁ parameters like germination, survival, seedling height, pollen and seed sterility showed positive significant

correlation with M_2 chlorophyll mutation frequency. Correlation between chlorophyll mutation and pollen and seed sterility was highest in both the varieties.

Population variance for seeds/pod, pods/plant and yield/plant in M_2 generation showed significant positive correlation with some of the M_1 parameters. A negative association between seedling height and seeds/pod in Dhauli and between seedling height and pods/plant in Nayagarh local was observed. In both the experiments, seed and pollen sterility showed significant positive correlation with M_2 variance.

M_3 GENERATION

Significant difference was recorded between treatments and between progenies within treatment. Significant increase in yield over control was observed in E_1 , E_2 , S_2 , N_1 , N_2 , S_2N_2 , E_1S_1 , E_2N_1 , S_2N_1 , E_1S_2 and S_1N_2 treatments of Dhauli and E_1 , E_2 , N_1 , N_2 , S_1 , S_2 , E_2S_2 , E_2N_2 , S_2N_2 , E_1S_1 , E_2N_1 , S_2N_1 , E_1N_2 and S_1N_2 of Nayagarh local. Within treatment variance was significant in 14 and 13 treatments of Dhauli and Nayagarh local respectively.

M_4 GENERATION

Significant difference was observed among all the cultures for all the 8 characters studied.

The estimates of PCV was moderate for plant height and pods/plant in both the varieties. The estimates of GCV followed similar trend and the values were less than their corresponding PCV. Heritability estimates were high (> 80%) in all the characters of both the varieties except 100-seed weight of Dhauli. In both the varieties the expected genetic advance was

high for pods/plant and plant height. High heritability along with high genetic advance was observed for pods/plant in both the experiments.

Significant correlation coefficient between yield and pods/plant, branches/plant and yield/plant was observed in Dhauri whereas, only between yield and pods/plant in Nayagarh local. The extent of association at phenotypic level was lower than the corresponding genotypic level in most pairs of characters. A non-significant association between some pairs of characters was also observed in both the varieties.

The path coefficient analysis in Dhauri revealed that direct contribution of pods/plant towards seed yield at genotypic level was highest followed by seeds/pod and 100-seed weight. In Dhauri the indirect effect of pods/plant via other characters tend to be small or often negative, as the direct effect of pods/plant was too high. The direct and positive contribution of seed weight was nullified due to indirect negative effect via pods/plant. Negative direct effect was observed for days to flowering and days to maturity. Though the strength of the direct effect of branches/plant was low, at genotypic level, its correlation with yield was enhanced due to positive indirect effect via other characters.

Among the mutant culture of Nayagarh local, the direct effect of pods/plant was the highest followed by 100-seed weight and seeds/pod, whereas, it was negative for days to flowering and plant height. The direct effect of 100-seed weight and seeds/pod to yield was nullified by negative effects via other characters resulting reduction on genotypic

correlation of this trait with yield.

Based on D^2 values all the 47 mutant cultures including control were grouped into 11 and 12 clusters in Dhauli and Nayagarh local respectively. The Z_1 and Z_2 graph using canonical analysis also confirmed to this findings. The clustering pattern of both the experiments indicated that cultures derived from one treatment often grouped into different clusters and vice versa.

The relative performance of different characters based on rank average indicated that 100-seed weight contributed maximum to the divergence in Dhauli. It was followed by days to maturity and branches/plant. In Nayagarh local, branches/plant contributed maximum to the divergence followed by days to maturity and plant height.

In both the experiments it was observed that cluster which included ~~containing~~ the parent yielded lower than other clusters. Cluster III was the highest yielder in Dhauli followed by Cluster XI. The cultures of these clusters had short height, medium maturity associated with high pods/plant, seeds/pod and moderate to high seed weight. In Nayagarh local, cluster XI had the highest yield followed by XII and VIII. Hybridization of genotypes having high D^2 values and differences in yield determining characters could produce transgressive variants for yield. Based on these, hybridization between DE-3 of cluster-IV and DE₂N₂-2 (Cluster-V) of Dhauli and NS₁-6 (Cluster-II) and NE₁S₁-12 from Cluster - VII may be taken up to obtain transgressive segregant.

Multiple selection criteria based on pods/plant,

seeds/pod and 100-seed weight (SC - II) was found the most efficient method of selection for the mutant cultures of Dhauli. It was followed by the criteria based on all the 8 traits (SC - V). For the cultures of Nayagarh local, the criteria selection based on pods/plant, seeds/pod, 100-seed weight and yield (SC - III) were found efficient over rest of the selection criteria in identifying desirable mutants.

Comparison of realised yield improvement and efficiency of five selection criteria indicated that selection indices like SC - II, SC - III and SC - V were more effective than direct selection on yield per se (SC - I). From the realised gain in mean yield of different selection groups in M_5 generation the relative performance of different selection criteria was assessed. In M_5 generation, the selected mutant cultures of both the varieties were significantly superior over the control with respect to yield and yield attributes. Further, these mutants were short in height and early maturing ones.

Conclusion :

It could be concluded that more chlorophyll mutants were observed in the treated populations of NG in both the varieties. A dose dependent response of mutagens were observed for different characters. Two varieties differed to mutagens with respect to production of macro- and micromutations. Thus, it could be better to treat different varieties differing in morphological traits for success from micro-mutational improvement for yield. Pollen and seed sterility percent in M_1 can be use as an index for predicting M_2 mutation frequency.

More numbers of elite lines were obtained from combine mutagenic treatments, particularly from EMS + SA combinations. Mutagenic line derived from mutagenic treatment could be grouped and genetically diversified micromutants from parent, could be identified for use in further breeding programme.

Selection from normal looking plants in M_2 , putting subsequent selection pressure in M_3 and use of selection criteria in M_4 could produce an efficient and effective method to select high yield micromutants.

CHAPTER - VI

SUMMARY AND CONCLUSION

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